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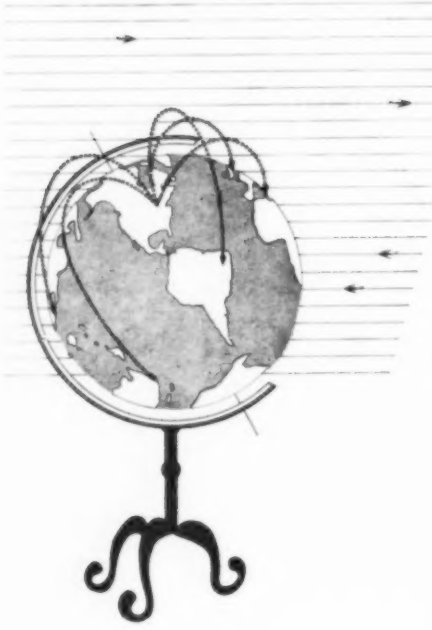

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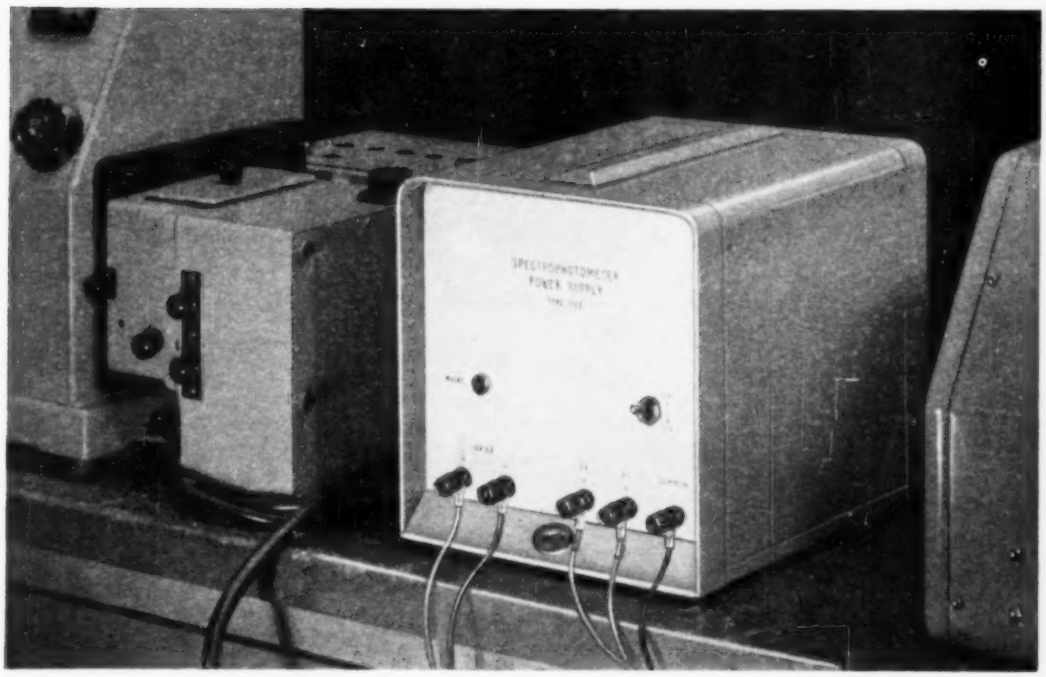

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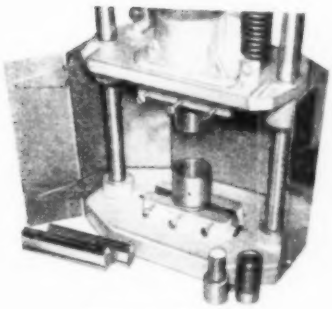

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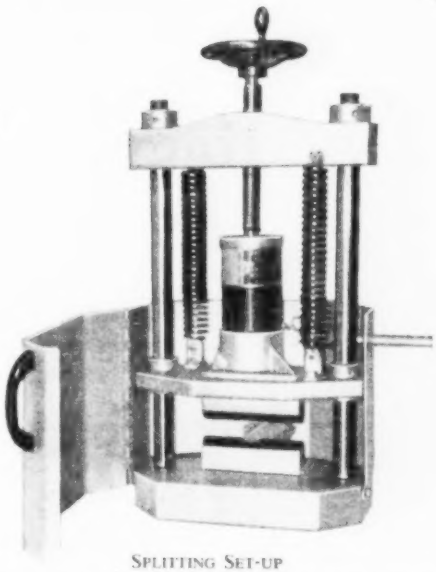

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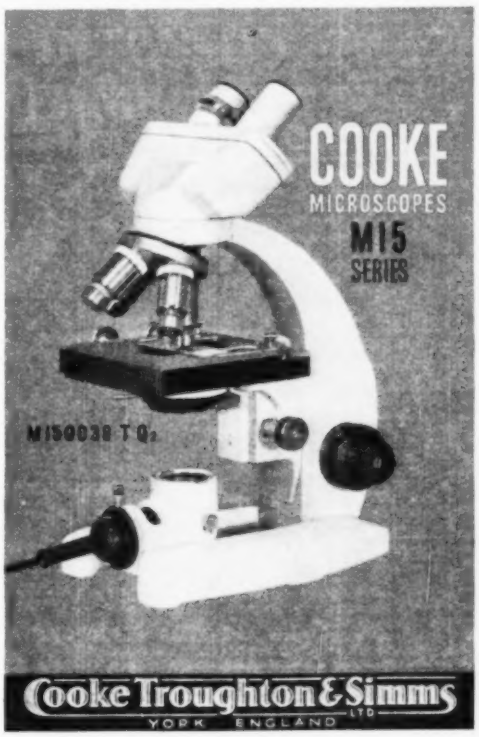

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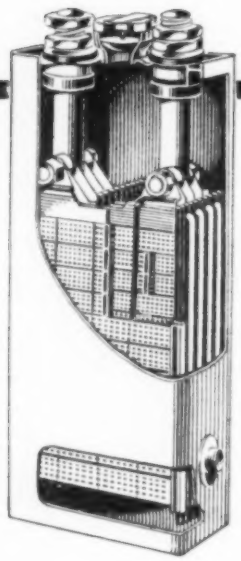

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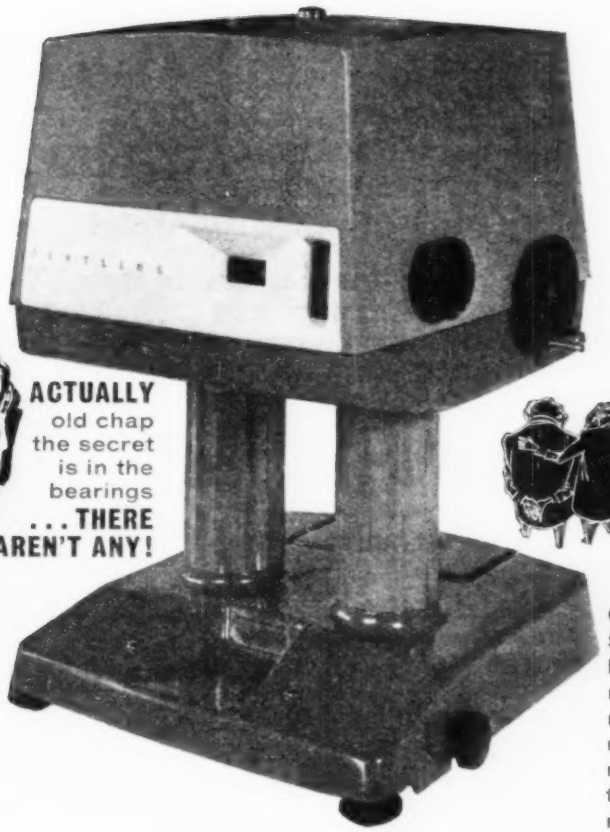

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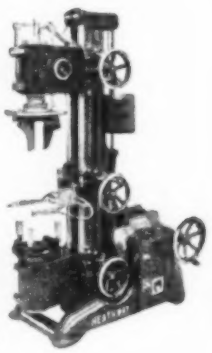

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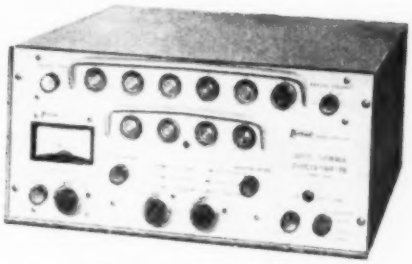

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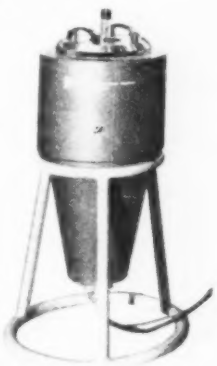

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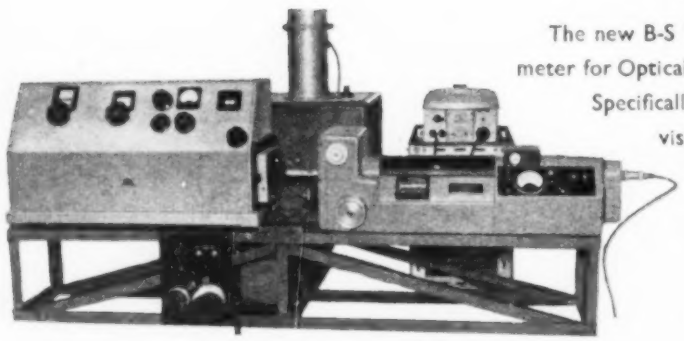

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"ALL this world is heavy with the promise of greater things, and a day will come, one day in the unending succession of days, when beings, beings who are now latent in our thoughts and hidden in our loins, shall stand upon this earth as one stands upon a footstool, and shall laugh and reach out their hands amidst the stars." So H. G. Wells predicted in his discourse, "The Discovery of the Future", delivered at the Royal Institution on January 24, 1902 (*Nature*, **65**, 326; 1902).

Almost sixty years later the Academy of Sciences of the U.S.S.R. has given point to that prophecy. Man is entering a new régime of endeavour. Risen from intertidal slime, he now cleaves interstellar plasma. The astronautical venture has begun. For those who, from innate scepticism or lack of knowledge, have underrated this epic achievement as being no more important than rocketing instruments into space, some guidance is necessary. Apart from the sheer poetry of a human being undertaking such a Ulyssean journey, the scientific value is vast. Instruments, however ingenious, can only yield what they have been set to do. They are precluded from the realm of discovery. Man, with his natural universality and ability to reason, is at the very pinnacle of all discovery.

Unfortunately, the perspective received by the public is a trifle confusing. The reason is that astronautical programmes have given the impression of being somewhat of a random process—not wholly untrue this side of the Iron Curtain. However, Albert Ducrocq, a savant of note in astronautics and cybernetics, has managed in his book, *Victoire sur L'Espace* *, to describe with much clarity the achievements and aims of space science. The book does not deal in detail with the development of space-vehicles, but provides an excellent review of performance and accomplishment. With Gallic limpidity of expression (not lost in the translation) he shows how the chief outcome of the astronautical revolution has been to make the scientific description of Nature more elegant.

Merely a few measurements made outside our planet have produced immense strides forward in certain branches of science. For example, as described in the famous report (March 28, 1959) by Academician A. N. Nesmeyanov, president of the Academy of Sciences of the U.S.S.R., the first *Sputniks* revealed that the density of the upper atmosphere is greater than was thought and composition is very different too, molecular forms of nitrogen and oxygen being essentially non-existent. Also, extreme turbulence with enormous variation in temperature and pressure appears to characterize this home of Vulcan. The very basis of meteorology has been transformed with such knowledge. Human intervention in directing the course of winds, rains and hurricanes may be envisaged. By analyses of the orbits, through application of Kepler's laws, the radius of the Earth has been estimated precisely. There appears to be a gravitational asymmetry suggesting a bulge in the southern part. Geophysical measurements have acquired a remarkable accuracy.

The existence of intense radiation at great height was indicated when the American *Explorer I* followed an eccentric orbit not planned by the technicians. It was an error on the plus side, since it led to identification, chiefly by Van Allen, of belts of radiation encircling our globe. It appears that the Earth is submerged within the Sun's atmosphere. These belts of plasma material having their origin in solar rays provide a link between solar events and their effects on Earth. For the first time outer space was used as a laboratory in an experiment known as 'Operation Argus'. An atom bomb was fired far out in space. Liberated electrons assembled almost instantaneously in the Earth's magnetic field to produce an artificial aurora. There is a danger in such experiments. Apart from causing the Earth's rotational speed to vary, repeated explosions could shatter the electrical equilibrium of the upper atmosphere, so playing havoc with our climates. This planet of ours seems to be rather delicately balanced.

What has distinguished the Soviet research in space is its direction toward the future. One investigation leads elegantly to another which, in turn, gives impetus to more extended work. While preparing for the future on a very large scale, Soviet scientists restrict the number of operations and make each as fruitful as possible. For example, it was puzzling that all three *Sputniks* should have been placed in orbits with low perigee, until one realizes that such satellites are at once atmospheric and geodetic, so yielding the greatest range of information. This powerful, ordered way of thinking is an interesting counterpoint to the impression one gets from the literature of Tolstoi, Dostoievsky and Bryúsov, for example, of the Russian mind, so passionately unrestrained. Therein probably lies their success.

Regrettably, one often finds naive dismissal of Soviet scientific achievement as being another Machiavellian device for propelling the propaganda machine and making overtures to the 'uncommitted'. To be sure, the Russians do relate political prestige to their scientific endeavour. But this should not compel us to denigrate the sheer scientific attainment itself.

M. Ducrocq discusses the categories of problems encountered in space travel and how some of these have been surmounted. First, stabilization of various kinds was required to prevent space-vehicles from tumbling. This was done by creating an initial spin or by using ancillary fluid-ejection devices. He reviews problems related to re-entry. When a satellite returns into the atmosphere, it takes up a streamlined attitude according to aerodynamic influences.

* The Conquest of Space: Moon Probes and Artificial Satellites—Their Impact on Human Destiny. By Albert Ducrocq. Pp. 300 + 8 plates. (London: Putnam and Co., Ltd., 1961.) 30*s.* net.

But, on crossing the interface, it is likely to experience tremendous oscillations, comparable with the flapping of a flag. Uncontrolled kinetic heating can have dramatic consequences, as in the case of the early V2's, which exploded, in mid-air, despite their relatively low speed. A human occupant of a space-vehicle has to contend with a range of gravitational effects, and, for interplanetary voyages, there is the hazard of the radiation belts.

In travel to the Moon or planets the traditional notion of a journey no longer applies, since both the point of departure and the destination are moving. Such a voyage must be conceived in four-dimensional space. An advance without precedent in the 'conquest of precision' occurred when *Lunik II* was navigated to reach the Moon. The *Sputniks* had attained a precision of 1 per cent. For a shot to the Moon this represents an uncertainty of several tens of thousands of miles (with a régime close to unity). In January 1959, *Lunik I* took off at an initial speed controlled to within 0·1 per cent, already a ten-fold increase in precision since the previous year. It passed at 4,660 miles from the Moon. In September 1959 the staggering success of a precision of 0·01 per cent was gained when the second lunar probe landed on its target.

Thus for the first time spatial guidance was introduced. Using an ingenious iterative technique designed by Prof. Kotelnikov and Prof. Chtchoukmo deviation from an ideal ballistic path is measured *en route*, then small corrections are made. The vehicle is virtually steered to the Moon, which subtends an angle of only 0·5 degree as seen from the Earth. This amazing feat of gaining on the average one decimal place per annum attests to the directing influence of the Soviet astronautical programme, namely, to combine power with accuracy, each step paving the way for its successor.

It seems scientists may have attained yet higher orders of precision; at present a Soviet missile is in hot pursuit of the goddess of sweet intimacy. Sadly, perhaps, she will have to reveal her mysteries. M. Ducrocq discusses possible voyages to Mars as well and provides a very good summary of the elements of ballistic theory in an appendix.

It is indeed wondrous to witness how mankind, though thwarted and frustrated, often by its very nature, can, like Ulysses, ever feel that force

"To follow knowledge like a sinking star
Beyond the utmost bound of human thought."

A FURTHER TRIUMPH FOR SCIENCE

AS is now universally known, American scientists and technologists achieved a great victory when, on May 6, they were responsible for the projection of a rocket carrying a man (Commander Alan Shepard) into sub-orbital space, the capsule being recovered intact with Commander Shepard alive and well. This is a record of a controlled flight 115 miles high and 302 miles down-range.

The congratulations of all must be extended to the American scientists and technologists who made this feat possible; and we express great admiration to Commander Shepard for his valour and devotion to duty.

This American achievement follows closely on that of the Soviet scientists, who, on April 12, succeeded in placing a controlled space-vehicle in orbit containing a man (Major Gagarin) and also recovered him alive and well (see *Nature* of April 22, p. 291).

In announcing their achievement, the Soviet political authorities stated: "We gladly place them [the scientific results] at the service of all peoples in the name of progress, happiness and the well-being of all human beings on Earth". Similarly, from the White House, the President of the United States, commenting on this most recent American space victory, stated: "Important scientific material has been obtained during this flight and this will be made available to the world's scientific community".

Here are two great scientific feats achieved by two Powers more or less diagonally opposed in political creed. While the world's scientists must share satisfaction in the almost fantastic progress thus revealed, all and sundry should view with grave concern and no little apprehension the politics behind it.

As pointed out in our previous comment (*Nature* of April 22), it still seems that only lip service is being paid to freedom for scientific research and, above all, freedom to publish and discuss scientific results—to the ultimate benefit of man. We are glad to read the Soviet and American promises to release these, their latest, results, but shall feel happier when such promises are implemented, for until then scientific progress is shackled and world peace possibly threatened. To-day, far too much science is censored, and this is bound to cause international bad faith and suspicion.

Commenting on the great American achievement, a spokesman in the Soviet Embassy in London said: "This stage of development was reached several years ago in the Soviet Union". Most men of science, and indeed others, will be prepared to accept this as a fact; but it is significant that the Russians, so far as we know, made no announcement at the time or published any scientific results afterwards. On the other hand, the Americans have not only published immediately the actual fact of their success, but also did everything they could (through radio, television, etc.) to allow the world to follow the last stages of their gigantic experiment (for there must have been many stages before the actual launching of the rocket)—even at the risk of enacting a failure.

It is relevant here to reveal that for some years *Nature* has tried to persuade the Soviet authorities (through the media of some of their scientists) to

allow their magnificent scientific results (not confined to space research) to be published for all to read; and very recently approaches have been made direct to political authorities in Moscow and to the Soviet Ambassador in London; but, alas, so far with negative results.

To most non-scientists, the successful pioneers in these recent space researches are Gagarin and Shepard. Yet, with all due respect to these brave men, they themselves were actually part of certain scientific equipment. The fundamental work was done by, and therefore praise is also due to, the scientists who made possible the heroic feats of these two men. In fact, these journeys into space are the results of the work of many people over many years, culminating in team work among scientists and non-scientists. Alas, however, thanks mainly to the fundamental defects in the world's means of mass information and education, the scientists are relegated by the layman to an almost intangible, certainly generally incomprehensible, position, and have to suffer such vulgar epithets as 'back-room boys' or 'boffins'.

Such men as Gagarin and Shepard have had, and will continue to receive, public acclaim and fêting which is their due, though much of this may well verge on hysteria stimulated by political propaganda and national patriotism. Meanwhile, the scientist goes quietly on with his work.

One sometimes wonders if such national and international acclaim would be accorded to a man who discovered an over-all cure for cancer or who made two blades of grass grow where only one or none grew before. Certainly no such world-wide publicity and praise were meted out to those scientists who were responsible for the discovery and development of penicillin in the late 'thirties and onwards; yet to-day, as a result of this work, millions of lives are being saved, much suffering alleviated and a great industry of antibiotics established in many parts of the world.

We would be the last to wish to denigrate the contributions Major Gagarin and Commander Shepard have personally made to the advancement of space science; but these two achievements, and especially the way in which they are being presented to the world, help to emphasize what *Nature* has been stressing continuously for some years, that is, the great danger inherent in allowing science to become the tool of political and diplomatic intrigue; for from scientific endeavour coupled with diplomatic intrigue it is only a short step to scientific success possibly coupled with political threat. Neither is it wise that scientific achievements be used for the enhancement of political prestige.

Until we have a world where all or most of its peoples are sufficiently educated or informed about things scientific, we shall travel further and further into that dread arena where politicians are playing ball with bombs—a dangerous game having no referee, not even the United Nations, who are certainly not yet wholly united in scientific endeavour.

MORE RECENT ADVANCES IN BIOCHEMISTRY

Recent Advances in Biochemistry

By Prof. T. W. Goodwin. Fourth edition. Pp. vi+301. (London: J. and A. Churchill, Ltd., 1960.) 50*s.* net.

IT will probably come as a surprise to most readers of this book to find it described as the fourth edition of *Recent Advances in Biochemistry*. The third edition of a book with this title appeared in 1931. It was written by Prof. J. Pryde and was a landmark in biochemical literature. The present book is by a different author and the whole subject of biochemistry has, of course, become transformed in the intervening thirty years. Two consecutive editions of a book could scarcely be more different; yet the present one is likely to take a similarly outstanding place in the literature. The aim of the book is to bridge the gap between the general textbook and the specialized review article, and to introduce senior undergraduate students and research workers to some of the present-day frontiers of biochemical discovery. In a book of eight chapters, it has been possible to deal only with selected topics, and emphasis has been placed on mechanisms of biosynthesis, with a leaning towards the chemical rather than the physiological aspects of biochemistry.

Chapter 1 deals with electron transport and oxidative phosphorylation. Here, as elsewhere in the book, the author describes not only the known facts but also their experimental basis. Chapter 2 is concerned with carbon dioxide fixation in higher plants and micro-organisms and in animal tissues. It includes a discussion on the Krebs urea-cycle. The chemistry and biosynthesis of phospholipids is covered in Chapter 3, and the biosynthesis of aromatic and heterocyclic ring compounds in the next two chapters. These are followed by a discussion of the nucleic acids: "The compounds which perhaps more than any dazzle biochemists and biologists alike with the glittering prospect of an explanation of the ultimate mysteries of living matter". The subject is treated from the synthetic rather than the functional aspect, but there is an interesting discussion of recent work on the infection of *E. coli* by different strains of bacteriophage. Chapter 7 deals with biosynthesis of terpenes and related compounds, and contains a detailed description of the steps leading to the biosynthesis of cholesterol. Carotinoids are included in this chapter, and here the author is on his home ground for he has contributed much to our knowledge of the biochemistry of these compounds. The book ends with a short but most valuable chapter on the regulation of cell metabolism, a subject of great interest and importance, but one which has scarcely reached the text-books yet.

Throughout the book there are numerous references to the more significant original articles and there is a useful index. The author makes very free use of abbreviations for the names of compounds: indeed, some pages look as if they might have been written in code. Some of these abbreviations are not in common use; others, such as DPN, may soon become obsolete (see *Nature*, **188**, 464; 1960). It would save readers a lot of unnecessary searching if, in future editions, an alphabetical key to abbreviations were supplied at the beginning or end of the book. The standard of production is good, and the numerous diagrams of formulæ and reaction schemes

are clearly set out. This book will be greatly valued by research workers and teachers and, perhaps above all, by undergraduates in their final year of honours biochemistry. Organic chemists will find much to interest them too. It is not easy reading because of the complexity of the subjects discussed, but the chosen ground could scarcely have been covered better in a book of this size. It is much to be hoped that Prof. Goodwin will have the time and patience to bring out further up-to-date editions of this book from time to time. D. C. HARRISON

ANIMAL BEHAVIOUR

Current Problems in Animal Behaviour

Edited by W. H. Thorpe and Prof. O. L. Zangwill. Pp. xiv + 424 + 2 plates. (Cambridge: At the University Press, 1961.) 45s. net.

THE phenomena of animal behaviour are being studied by a rapidly increasing number of scientists. Few other biological phenomena have attracted such a variety of workers and have led to so many differently oriented disciplines. After a period in which these various disciplines ignored each other, a tendency grew towards establishing contact; this at first expressed itself in mutual criticism, but it is gradually developing into real co-operation. Because the subject is so complicated, because it also offers more obstacles to consistent scientific inquiry than most other life processes, and further because each class of workers has developed its own concepts, terminology and theories during periods of isolation, the growth of co-operation is a difficult and painful process. In spite of this, and notwithstanding a growing tendency towards specialization, an increasing number of workers is seriously attempting to build at least a common foundation for a unified, broad and systematic attack on problems of animal behaviour. These attempts naturally require much discussion, and in many research centres there have been formed interdisciplinary discussion groups in which problems, methods, concepts and concrete research plans are critically considered.

The present book gives us an insight into the kind of activities of such seminars. Each member of Dr. Thorpe's and Prof. Zangwill's alert and able group "was asked to write an essay on an aspect of his own work or discipline which appeared to him interesting and vital for the study of behaviour as a whole"; these essays were then critically discussed and rewritten.

The result is a series of extremely worth-while chapters, loosely bundled into four major parts. The editors jointly wrote a general introduction, and each part is again introduced by one of them.

Part 1 ("Neural Mechanisms and Behaviour") contains a chapter by D. M. Vowles in which differences between the nervous systems of insects and vertebrates are traced, and constructive chapters by L. Weiskrantz and O. L. Zangwill. Part 2, not quite appropriately called "Experimental Studies of Animal Behaviour", contains chapters on ritualization (A. D. Blest), on flock organization in birds (J. H. Crook) and on filtering of external stimuli (P. Marler); Part 3, on "Problems common to the Psychology of Animals and Men", has 5 chapters (R. A. Hinde, W. H. Thorpe—whose contributions overlap a little—M. A. Vince, D. E. Broadbent and A. J. Watson) and the last part deals with "Theoretical Approaches" (R. L. Gregory, H. B. Barlow and V. Cane).

While such a book can of course not be expected to be an exhaustive review, it is amazing how many aspects of the field are given critical attention, and how many chapters present really new points of view. Not unexpectedly, there is a great deal of trial-and-error thinking in print, and if the trend of some contributions is a little over-destructive, this reflects the soul-searching state our science is in. Nor should it be forgotten that there has been a considerable delay in publishing.

The title, and particularly the sub-title, perhaps suggest a more exhaustive covering and more unity of approach than are really presented. When the editors state that "the psychologist is . . . interested in . . . the explanation of behaviour in general" (p. x) they are, I feel, underrating the facts that the narrow descriptive basis of psychology makes it at times difficult to distinguish the general from the special, and that the undoubtedly general problems of survival value and evolution are still neglected in psychology. Furthermore, when they say that "biology is *still* very largely dominated by the concept of evolution" (p. xii, italics mine), do they mean that this is a passing stage, which biology will outgrow, and which psychology has already outgrown? N. TINBERGEN

AXIOMATICS AND FOUNDATIONS

The Axiomatic Method, with special reference to Geometry and Physics

Proceedings of an International Symposium held at the University of California, Berkeley, December 26, 1957–January 4, 1958. Edited by Prof. Leon Henkin, Prof. Patrick Suppes and Prof. Alfred Tarski. (Studies in Logic and the Foundations of Mathematics.) Pp. xi + 488. (Amsterdam: North-Holland Publishing Company, 1959.) 90s.

The Foundations of Mathematics

A Study in the Philosophy of Science. By Evert W. Beth. (Studies in Logic and the Foundations of Mathematics.) Pp. xxvi + 741. (Amsterdam: North-Holland Publishing Company, 1959.) 100s.

THESE two books, different in emphasis but alike in purpose, present an inspiring picture of modern thought concerning the basis of mathematics. The drilling into the sub-structure of the subject goes on with relentless vigour. The whole field is becoming so vast that expertize within expertize is beginning to appear, which means that the student needs instruction of course, but guidance as well, through the mass of highly specialized literature. All this is provided in good measure.

Axiomatic Method is the outcome of a conference held in 1958. Its general theme, as the title suggests, is the formal structure of mathematics. The deliberations are divided into three parts: (1) foundations of geometry; (2) foundations of physics; (3) general problems; and applications of the axiomatic method. In the first of these the order is that of subject,

in the second of historical development, and in the third (broadly) from the more specific to the more general. The reader will appreciate this plan, but the purpose behind the arrangement is worth noting. Geometry has been the home ground of axiomatics for more than two thousand years, and in that field the method originated. It would be quite unnatural if any sequence other than by subject were adopted. But physics has not developed like that, and so the chronological treatment is the better. In fact, to-day physical research is often curiously informal (in the technical sense), and its theoretical side is frequently kept lively and progressive by the need to resolve some inconsistency or to remove some apparent paradox thrown up by experiment. Newton's *Principia* has indeed more than a touch of Euclidian methodology. But of that, more anon.

There are at least two classic examples in which the axiomatic principle worked well in physics under the hand of a great master. The first was Archimedes, and the second Carathéodory, who in 1908-9 set up a system of thermodynamics on the basis of two axioms. Neither of these receives comment. The omission is by no means serious, since much of the work discussed is—very properly—modern. But it would have been interesting to have had the editors' reactions to the contributions made by such outstanding minds separated by such a long period of time.

Concerning the particular 'atmosphere' of the Newtonian period, and the seeming lack of postulates or any other kind of 'common notions', it looks very much as if the influence of Francis Bacon was too dominant, and that stress was laid very firmly on the power of science (in general) as an instrument of progress. If this should be a correct interpretation of events, it would follow that induction rather than deduction was *de rigueur* in the prevailing intellectual climate.

Turning to applications of the axiomatic method (Part 3), Prof. R. L. Wilder, of the University of Michigan, has a most intriguing paper on the innovations which he brought about in teaching his postgraduate students to use such a powerful tool for themselves in the establishment of proofs, sufficiently refined and complete to satisfy twentieth-century standards of consistency and adequacy. In this way (and occasionally even at undergraduate-level) he claims to have discovered creative talent otherwise dormant or perhaps frustrated.

The Foundations of Mathematics has a somewhat different aim. It is essentially a treatise, covering almost all aspects of foundation theory, but axiomatic in spirit, though this aspect is confined to Parts 2 and 3. Part 1 provides an exceptionally lucid study of the historical background of foundations, with particular reference to Aristotle's Theory of Science, and the rise of symbolic logic—which must, almost of necessity, include a brief outline of formalized axiomatics. Further parts deal with such topics as non-elementary mathematics, the existence of mathematical entities, paradoxes, set-theory and topology, recessive functions, sets and predicates. In conclusion come some comprehensive remarks on metamathematics and philosophy. Prof. E. W. Beth has some shrewd observations to make on the countless—often amateurish—efforts to resolve the more important paradoxes. Having expounded the orthodox views on such matters, he implies, very gently, that they have too often suffered from the 'fools and angels' type of attention. It is all too easy to under-estimate the methodological significance of the paradoxes, as well as to fall short of present-day standards of rigour in any approach to clarify their more intractable features.

The author's method is, in principle, analytic. He is thus enabled to deal with questions as they arise, rather than to ask his readers to accept assertions which may be far from obvious. Again, he recognizes that even logic may be a matter of degree: beginning at an "average level", he is able to scale the heights as the work proceeds. From the students' point of view, the realization of this dynamic process is important.

Reference to sections would be facilitated if the section numbers could be printed at the top of each page. As it is, much searching is involved to find such necessary information, especially when an equation or formula is first encountered, and then mentioned again many pages ahead.

F. I. G. Rawlins

GROUPS, PURE AND APPLIED

The Theory of Groups

By A. G. Kurosh. Translated from the Russian and edited by K. A. Hirsch. Second English edition. Vol. 1: Pp. 272. Vol. 2: Pp. 308. (New York: Chelsea Publishing Company, 1960.) 4.95 dollars each volume.

Abelian Groups

By L. Fuchs. Pp. 367. (London and New York: Pergamon Press, 1960.) 63s. net.

Seminar on Transformation Groups

By Armand Borel. With contributions by G. Bredon, E. E. Floyd, D. Montgomery and R. Palais. (Annals of Mathematics Studies, No. 46.) Pp. viii + 245. (Princeton, N.J.: Princeton University Press; London: Oxford University Press, 1960.) 36s. net.

THE two volumes by A. G. Kurosh, in their American translation by K. A. Hirsch, as well as the book by L. Fuchs, are photographic reprints of the first editions that appeared in 1955–1956 and 1958 respectively. A few misprints have been corrected in this second American edition of Kurosh's book; the Pergamon Press has appended a short list of errata to Fuchs's book which was first published by the Hungarian Academy of Sciences, but the present reprint is not even described as a second edition. The short space of time that has elapsed since their first appearance is a measure of the success and the importance of these books. But a glance at their history will explain why this time is too long for them to re-appear unchanged: it would have been more in keeping with the spirit of these books if some of the recent advances that they themselves have done so much to stimulate had been included.

The theory of groups, now one of the most important branches of abstract algebra, has seen a spectacular burst of activity during the past few decades. Kurosh's book is remarkable for the way in which its various editions are in turn result and cause of this development. Kurosh first wrote *The Theory of Groups* in 1940. It was then the first book on just groups, rather than finite groups, and the stress was on infinite groups. The War delayed publication

until 1944, and the book became available to a wider public only in 1953 when the Akademie-Verlag, Berlin, published a German translation. By that time the stimulus provided by the Russian original had led to numerous new discoveries, some of which were added to the German translation in an appendix by B. H. Neumann. In the meanwhile, Kurosh himself had completely recast and revised his book in the light of the rapid advances, especially in the theory of infinite nilpotent groups, made by the powerful school of Russian group theorists. Thus, in the same year 1953, the second Russian edition appeared, virtually a new book compared with the first edition. It is from this second edition that Hirsch's translation was made. Once again Hirsch supplemented the translation by notes reporting on more recent developments. He also brought the bibliography up to date—that is, up to 1956—so that it now includes more than 800 titles.

The theory of abelian groups, though part of general group theory, has its own methods and its own history of growth. A first account of the theory of infinite abelian groups appeared in 1954, in a well-known monograph by I. Kaplansky. Here again further progress was rapid, and Fuchs's book in 1958 came none too early. Naturally, so far as the basic structure theory is concerned the books overlap. Beyond that much of the characteristic flavour of Fuchs's book is due to the fact that it, too, comes from the pen of one of a lively school of research workers in the subject, this time the Hungarian school of group theorists. Indeed, a glance at the extensive bibliography and at the numerous examples at the end of each chapter, many of them provided to give additional information on recent results in concise form, reveals the vigour of the (then) current research.

Both books start from the beginnings of the subject, assuming no more than a certain maturity of mathematical reasoning and familiarity with some basic mathematical tools. Both lead to the frontiers of knowledge in their time, giving not only the latest results but also pointing again and again to the unsolved problems and open questions. Thus both books are suitable for, in fact strongly recommended to, the bright undergraduate and the young research student; at the same time their depth and scope make them indispensable for the expert. Some of the most striking discoveries made since their appearance owe much to the influence of these books; many of the problems they state are, in fact, now solved. Let us hope that a further edition may soon be possible.

By contrast, the book by Armand Borel and his collaborators deals with a highly specialized application of group theory and homology theory. It embodies the results of a seminar held by Borel at the Institute of Advanced Studies at Princeton, N.J.; several chapters are written by members of the seminar presenting their own contribution to its work. The topic is the application of cohomological methods to the study of the action of compact transformation groups on topological spaces; the tools, ranging far and wide over recent work in algebraic topology, are mainly taken for granted, though a general reference to the sources is provided. At this different level of specialization the presentation is again clear and well organized, but naturally concise. Each chapter carries its own short bibliography, not aiming at completeness but containing only the most relevant papers. This book is clearly for the expert only.

HANNA NEUMANN

POWDER METALLURGY

Fundamental Principles of Powder Metallurgy

By Dr. W. D. Jones. Pp. xii + 1032. (London: Edward Arnold (Publishers), Ltd., 1960.) 147s. net.

POWDER metallurgy is becoming an increasingly important metallurgical process, and, though a number of comprehensive books on the subject have been published in the United States, it is very useful to have this long and detailed book published in Great Britain and written by an author with such wide experience in powder metallurgy. Dr. Jones is the author of the well-known book *Principles of Powder Metallurgy* published in 1937.

The new book starts with a long chapter of 239 pages on the preparation of powders and then has three chapters, totalling 350 pages, on "Pressing", "Shaping without Pressing" and "Sintering". These are followed by another very long chapter on "Attainment of Specific Qualities", which deals mainly with the properties required in products made by powder metallurgy, but which also contains a section on factory management. The book ends with two short chapters on "Continuous Powder Metallurgy" and "Methods of Control"; the former covers the three main methods of continuous compacting of powders; the latter is very short and merely lists the various methods of testing powders and compacts and states where details of the actual tests can be found.

The author states in his introduction that he intends to cover principles rather than practice and so detailed descriptions of plant and processes are not included. Apart from this omission the book is very comprehensive; very little, if anything, appears to have been left out. The book is written in a clear and simple manner with good and relevant illustrations.

The book will be essential for all metallurgical libraries, since some at least of the matters described will be of importance or interest to all metallurgists. It will also be useful to others who are not metallurgists, so important are powders in modern technology.

It is possible that with the development of processes such as the Sherritt–Gordon process for the reduction of metals from their ores, the final products of which are powders, powder metallurgical products will no longer be used only for small or specialized components but will also be used for large-scale processes.

As has been said, the book is long, more than a thousand pages, and at seven guineas is expensive. The length and cost could have been reduced by omitting parts which do not fall strictly within the province of powder metallurgy or which can be found in standard text-books. Examples are the descriptions of the properties of metal whiskers and of general magnetic properties. The section on "Powder Metallurgy as a Competitive Process" does not, as might be expected from its title, show when it is economical to use powder metallurgy rather than other processes, but describes the organization of a factory producing powder metallurgy products. The section is interesting, but it is doubtful whether the organization of such a factory differs in its essentials from any other factory producing small parts.

J. H. RENDALL

The Brittle Fracture of Steel

By Dr. W. D. Biggs. Pp. xxvi+420. (London: Macdonald and Evans, Ltd., 1960.) 70s. net.

IN this book the author gives a very thorough account of all the factors related to the brittle fracture of structural steel. Thus he details a vast amount of accumulated knowledge on deformation and fracture, and discusses service and design aspects as well as methods of testing and the effect of metallurgical variables on behaviour. In addition, he deals with the relation between welding and brittle fracture, and the significance of residual stresses. Although cast irons are briefly mentioned, the coverage on steel does not extend to alloy steels as such.

The text is authoritative and comprehensive. However, as a result of the wide treatment the book does not readily yield a concise overall picture. The diffuseness in increased by the fact that in places the style is rather involved. This tendency is most noticeable when the author is generalizing rather than when he is giving specific information. There is indeed a useful summary chapter of fourteen pages, but it would have been better if this had been subtitled and set down in a crisper style.

In conclusion, it must be emphasized that this is a sound and informed book providing a thorough development of the subject-matter, and in this respect it deserves recognition as a valuable reference work. Regarding the style, the remarks here of course merely indicate my own reaction. Nevertheless, it does seem that the book would have been much easier to assimilate had the author cut through the subject in a more incisive manner.

A. R. BAILEY

Proceedings of the Fourth International Conference on Ionization Phenomena in Gases

Uppsala, 17–21 August, 1959. Edited by N. Robert Nilsson. (Series in Physics.) Vol. 1: Pp. xii+552. Vol. 2: Pp. viii+553–1210. (Amsterdam: North-Holland Publishing Company, 1960.) £12 6s. 0d. the two volumes.

THESE two volumes contain the majority of the 250 papers which were read at the conference by scientists actively engaged in the field of ionization physics in twenty-one countries throughout the world. The papers, by far the greater number of which are in English, cover a very wide range of topics, but they have been conveniently grouped so that Volume 1 contains those concerned with fundamental ionization processes and various types of gas discharges, and Volume 2 those dealing with various aspects of plasma physics. (More detailed information about the topics discussed under these general headings was given in a recent report on the conference[1].) It would have been helpful to the reader if the review papers, given to the plenary sessions of the conference, could have been distinguished in some way from the more detailed papers read to the four parallel sessions into which the conference was divided for much of the time. It is also a pity that the often lively and informative discussion of the papers had to be omitted for reasons of space. These, however, are minor criticisms, and both the editor and publishers are to be congratulated on providing such a very well-produced record of this important and extremely well-organized conference. All those with an interest in this field of work will wish to have ready access to these volumes in their departmental library, even if the inevitably high price precludes them from owning their own personal copies.

J. DUTTON

[1] Dutton, J., Harcombe, D., and Jones, E., *Nature*, **184**, 1353 (1959).

Determination of the Mechanical and Technological Properties of Metals

By B. M. Gliner. Translated from the Russian. 2nd edition. Translation Editor: E. Bishop. Pp. ix+160. (London and New York: Pergamon Press, 1960.) 50s. net.

THIS book gives a useful résumé of Russian mechanical testing methods. On the assumption that it contains a reasonably modern exposition (no date is given in the translated preface) of the varied techniques, it reveals no startling divergences from Western practice, although there are two unusual features that are mentioned.

One of these features is the use of ring and split-ring test pieces, for testing under short-time transverse tension, at room temperature, and transverse creep tension, at elevated temperature, respectively. It is not made clear why, or on what scale, such test pieces are used, or what information, other than an empirical guide to design, is gained from the tests.

The other unusual feature is the use of a triangular pyramid indentor for micro-hardness testing. It is difficult to see what particular advantage is gained by this form of indentor, other than to average out directionality effects in the material.

A statement is made (on page 14) that load measurement is most accurately achieved by means of a pendulum lever mechanism. This will certainly be questioned by Western manufacturers unless it is taken as a reflexion on Russian machine construction methods.

There are one or two minor errors to be found; for example, Fig. 61(*b*) is incorrect.

If the translation were less literal then the book would be easier to read. For example, on p. 16, "jockey" would read better than "trolley", on p. 17, "drum" would be better than "box", and "I-section" would be better than "Type 1", and on p. 93, "disk" is preferable to "shaft".

JAMES G. TWEEDDALE

Practical Heredity with Drosophila

By Dr. Gordon Haskell. Pp. xii+124. (Edinburgh and London: Oliver and Boyd, Ltd., 1961.) 10s. 6d. net.

IT is surprising that this useful little book has not been written before. Elementary practical genetics is studied in many schools and colleges, and, because of inexperience of the manipulators, many breeding errors occur which could have been avoided. Haskell's book provides valuable hints on the means of obtaining and rearing pure stocks of *Drosophila*, the apparatus required to do this and the right ways to use it, the selection of suitable mutants to demonstrate genetic principles, statistical tests for evaluation, the cytology of *Drosophila* and an introduction to biometrical investigation. Many workers using *Drosophila* who have tried in vain to mate already pregnant females will be glad of all the practical help which Haskell has compressed into quite a short space.

T. H. HAWKINS

SEASONAL VARIATION AND AGE OF RADIOACTIVE FALL-OUT

By R. P. PARKER and J. O. CROOKALL

Physics Department, Institute of Cancer Research, Royal Cancer Hospital, Fulham Road, London, S.W.3

A NUMBER of observers in the temperate zones of the northern hemisphere have reported a seasonal variation in the concentration of long-range fall-out material[1–3]. Thus both dust samples collected from surface air and rain-water samples have shown higher activities in the late spring than in the late autumn. It has been suggested by Martell[4] that the spring maxima are due to the descent of fall-out material injected into the atmosphere by tests in the previous autumn, while Stewart *et al.*[1] have suggested an explanation on meteorological grounds using the Brewer–Dobson model of atmospheric circulation[5,6]. On the latter thesis the cold air which forms above the Pole during the late winter sinks in the spring, thus bringing down stratospheric air and associated fission debris. More recently Feely and Spar[7] have postulated that the movement of fission debris from the stratosphere into the troposphere primarily occurs by a process of turbulent mixing in the region of the troposphere gap.

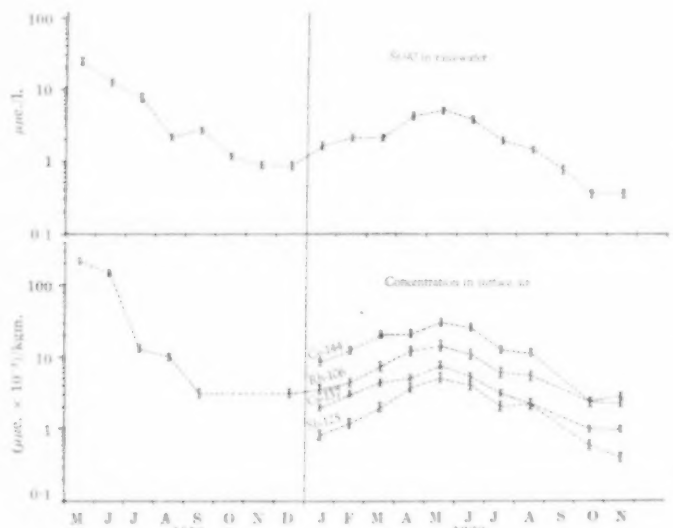

Fig. 1

In the past it has been difficult to assess the relative importance of these views owing to the periodic injection of fresh nuclear debris into the atmosphere. For example, Anderson *et al.*[3] have shown that while the fall-out pattern for 1959 was consistent with Martell's suggestion, it could not be taken to exclude a seasonal variation of the type suggested by Stewart. The spring of 1960 presented the first opportunity of studying this variation virtually free from the effect of recent tests. (The tropospheric fall-out from the 1960 French tests had only a transitory effect in Britain[8], and the yields were too small to produce a significant contribution to the total activity in the stratospheric reservoir.) We have already reported measurements for the early part of 1960[2] which indicate the beginning of a rise in the concentration of fall-out material, and more recently Peirson *et al.* have published measurements of the caesium-137 content of surface air up to the middle of 1960, which show a maximum in the spring[3]. We present here measurements of the principal γ-emitting fission products in surface air in 1960, together with values for the strontium-90 content of rain-water during this period. As before[2,9], samples of dust collected from surface air by means of an electrostatic precipitator at our laboratory at Sutton, Surrey, have been analysed by means of γ-spectrometry.

The principal γ-emitting fission products present in our 1960 samples are cerium-144, caesium-137, rhodium-106 and antimony-125, as expected for a period 12–24 months after fission[10]. In addition to the photo-peaks in the γ-spectrum which may be ascribed to the above nuclides, the identity of which has been corroborated by decay measurements on a number of samples, a peak occurs at an energy of 0·475 MeV. We have previously shown[9], on the basis of chemical and physical evidence, that this is in part due to 53-day beryllium-7, a product of the interaction of cosmic rays with atmospheric nitrogen and oxygen nuclei. The situation is now further com-

Table 1. Concentrations (curie × 10^{-12} per kgm. air) of γ-Emitting Fission Products in Surface Air

Mean date of collection	Period of collection (days)	Rh-106	Sb-125	Cs-137	Ce-144	Ce-144 / Cs-137
2.1.60	22	3·2	0·8	1·7	7·5	4·4
30.1.60	8	4·6	0·9	2·7	11	4·1
17.2.60	10	5·0	1·3	3·2	13	4·1
4.3.60	6	6·2	1·9	3·9	16	4·1
18.3.60	22	7·4	2·1	4·8	21	4·4
3.4.60	10	11	3·2	5·4	25	4·6
14.4.60	13	12	3·4	5·7	22	3·9
23.4.60	5	13	4·4	7·2	27	3·8
29.4.60	6	14	6·2	8·1	34	4·2
9.5.60	8	17	6·1	8·1	35	4·3
21.5.60	7	15	4·2	7·1	29	4·1
4.6.60	5	20	6·4	12	48	4·0
14.6.60	14	8·9	4·4	5·2	22	4·2
29.6.60	15	8·9	2·6	5·4	18	3·3
14.7.60	15	5·2	1·9	3·9	11	2·8
1.8.60	8	5·5	2·0	3·7	12	3·2
15.8.60	7	5·6	2·4	3·1	11	3·5
11.10.60	13	2·4	0·6	1·0	2·4	2·4
9.11.60	14	2·3	0·4	1·0	2·8	2·8

Table 2. CONCENTRATION (μμC/L.) OF STRONTIUM-90 IN RAIN-WATER SAMPLES, 1960

Period of collection	Sr-90
January	1·7
February	2·2
March	2·2
April	4·4
May	5·3
June	3·0
July	2·0
August	1·5
September	0·79
October	0·42
November	0·46

plicated by the presence of 210-day rhodium-102, recently reported to have been produced as a tracer in certain of the American 'Hardtack' explosions of April–August 1958[11,12]. Recently this nuclide has been stated to emit principal γ-rays of energy 0·475 MeV. (63 per cent) and 1·09 MeV. (6 per cent)[13]; but earlier decay schemes suggested a higher intensity for the 1·09-MeV. line. The rhodium-102 photo-peak at 0·475 MeV. cannot be resolved in our apparatus from the 0·478-MeV. line of beryllium-7, and, if we accept the new data, the intensity of the 1·09-MeV. line is too weak for it to be used in the identification of the small amounts of rhodium-102 which may be present in our samples. We are therefore carrying out further work to determine the relative amounts of beryllium-7 and rhodium-102.

Our measurements on the specific concentrations of rhodium-106, antimony-125, cæsium-137 and cerium-144, corrected for decay to the time of the collection, are given in Table 1. After averaging over calendar months, the results are plotted in Fig. 1, together with the strontium-90 concentrations in rain-water samples (Table 2). These measurements form a continuation of work previously reported[2].

The common feature of all these results is the pronounced rise to a maximum in May, with a fall during the summer months to low autumn and winter values. From June until October the values for the strontium-90 concentrations in rain are a close fit to an exponential curve of half-period 40 days, which is remarkably similar to our results for 1959[2]. The ratio between maximum and minimum in 1960 is approximately 5 : 1. This is in agreement with results for strontium-90 in rain during 1955–58[14], and with the finding that the poleward transfer of tungsten-185, liberated as a tracer in certain of the American tests near the equator, is seasonal[7,15]. It would therefore appear that our results may be explained in meteorological terms by the Brewer-Dobson or Feely–Spar models, since the Martell thesis is not applicable in 1960.

Information on the age of the debris collected may be obtained from the relative proportions of the various nuclide activities present in our samples. The yield of both cerium-144 and cæsium-137 is relatively independent of the type of fission process (Table 3)[16], and therefore the ratios of the specific activities of these nuclides provide a suitable method of determining age at the present time. The concentrations of nuclides of shorter half-life, such as strontium-89 and zirconium-95, are now too low for accurate measurement[3].

Table 3. FISSION YIELD (PER CENT) FOR DIFFERENT TYPES OF FISSION (ref. 16)

Mass-chain	Half-life	U-235, n_f	Pu-239, n_f	U-238, n_f	U-238, n_{14}	U-235, n_{14}
106	370 days	0·47	6·17	2·61	3·11	1·40
125	2·4 years	0·32	0·26	0·05	1·00	1·36
137	30 years	6·18	6·14	6·45	5·71	5·10
144	285 days	4·76	3·66	4·42	3·32	3·30

n_f, fission spectrum neutrons; n_{14}, 14-MeV. neutrons.

Our experimental cerium-144/cæsium-137 ratios are listed in Table 1, and it is interesting to note that during the early part of 1960 they are in good agreement with values quoted for rain-water at Milford Haven[16]. Using the theoretical yield values (Table 3) the average age for each sample has been calculated, and the results are presented in Fig. 2. The error in any one value is estimated to be ± 20 per cent. If we take the mean of these estimated dates of origin, a time April–May 1958 is obtained.

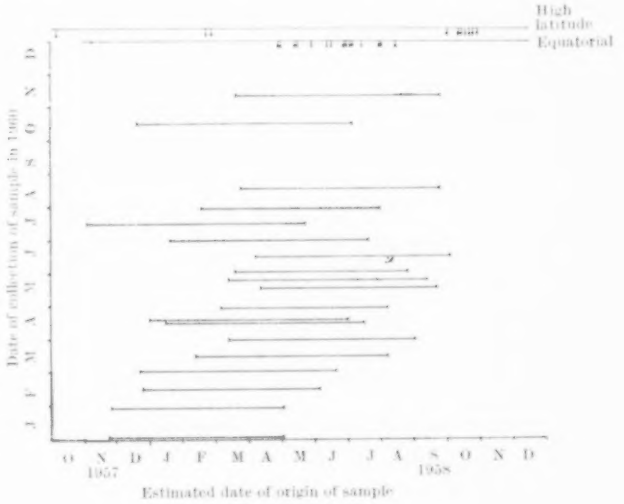
High latitude
Equatorial
Date of collection of sample in 1960
J F M A M J J A S O N D
O N D J F M A M J J A S O N D
1957
1958
Estimated date of origin of sample

Fig. 2

This is compatible with available information on the dates of megaton explosions[17]; these are indicated by arrows at the top of Fig. 2. The above calculation is based on the fission yield for 14-MeV. neutrons, and in the unlikely event of all the yield being due to the fission of uranium-235 by fission neutrons the mean date of origin would be approximately four months earlier. Thus the majority of fall-out material collected at our site during 1960 probably originates from the American and British equatorial tests of 1958.

The antimony-125/cæsium-137 and rhodium-106/cæsium-137 ratios had experimental values in mid-1960 of approximately 0·6 and 1·9 respectively. These are commensurate with an age of 3–4 years if 14-MeV. fission yields are used. However, the yield of both antimony-125 and rhodium-106 is very sensitive to the type of fission process, as shown by Table 3. Thus the discrepancy may be explained if, in addition to the 14-MeV. fission of uranium-238, other fission processes are involved.

We wish to acknowledge the guidance and encouragement of Prof. W. V. Mayneord and Dr. W.

Anderson during this work. We also wish to thank Mr. R. W. Kent and Mr. D. Smith for their help in connexion with the electrostatic precipitator, and Mrs. A. Perry for assistance in the chemical analyses.

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AN UNSTABLE INTERMEDIATE CARRYING INFORMATION FROM GENES TO RIBOSOMES FOR PROTEIN SYNTHESIS

By Dr. S. BRENNER
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A LARGE amount of evidence suggests that genetic information for protein structure is encoded in deoxyribonucleic acid (DNA) while the actual assembling of amino-acids into proteins occurs in cytoplasmic ribonucleoprotein particles called ribosomes. The fact that proteins are not synthesized directly on genes demands the existence of an intermediate information carrier. This intermediate template is generally assumed to be a stable ribonucleic acid (RNA) and more specifically the RNA of the ribosomes. According to the present view, each gene controls the synthesis of one kind of specialized ribosome, which in turn directs the synthesis of the corresponding protein—a scheme which could be epitomized as the one gene–one ribosome–one protein hypothesis. In the past few years, however, this model has encountered some difficulties: (1) The remarkable homogeneity in size[1] and nucleotide composition[2] of the ribosomal RNA reflects neither the range of size of polypeptide chains nor the variation in the nucleotide composition observed in the DNA of different bacterial species[2,3]. (2) The capacity of bacteria to synthesize a given protein does not seem to survive beyond the integrity of the corresponding gene[4]. (3) Regulation of protein synthesis in bacteria seems to operate at the level of the synthesis of the information intermediate by the gene rather than at the level of the synthesis of the protein[5].

These results are scarcely compatible with the existence of stable RNA intermediates acting as templates for protein synthesis. The paradox, however, can be resolved by the hypothesis, put forward by Jacob and Monod[5], that the ribosomal RNA is not the intermediate carrier of information from gene to protein, but rather that ribosomes are non-specialized structures which receive genetic information from the gene in the form of an unstable intermediate or 'messenger'. We present here the results of experiments on phage-infected bacteria which give direct support to this hypothesis.

When growing bacteria are infected with a virulent bacteriophage such as *T2*, synthesis of DNA stops immediately, to resume 7 min. later[6], while protein synthesis continues at a constant rate[7]. After infection many bacterial enzymes are no longer produced[8]; in all likelihood, the new protein is genetically determined by the phage. A large number of new enzymatic activities appears in the infected cell during the first few minutes following infection[9], and from the tenth minute onwards some 60 per cent of the protein synthesized can be accounted for by the proteins of the phage coat[7]. Surprisingly enough, protein synthesis after infection is not accompanied, as in growing cells, by a net synthesis of RNA[10]. Using isotopic labelling, however, Volkin and Astrachan[11] were able to demonstrate high turnover in a minor RNA fraction after phage infection. Most remarkable is the fact that this RNA fraction has an apparent nucleotide composition which corresponds to that of the DNA of the phage and is markedly different from that of the host RNA[11]. Recently, it has been shown that the bulk of this RNA is associated with the ribosomes of the infected cell[12].

Phage-infected bacteria therefore provide a situation in which the synthesis of protein is suddenly

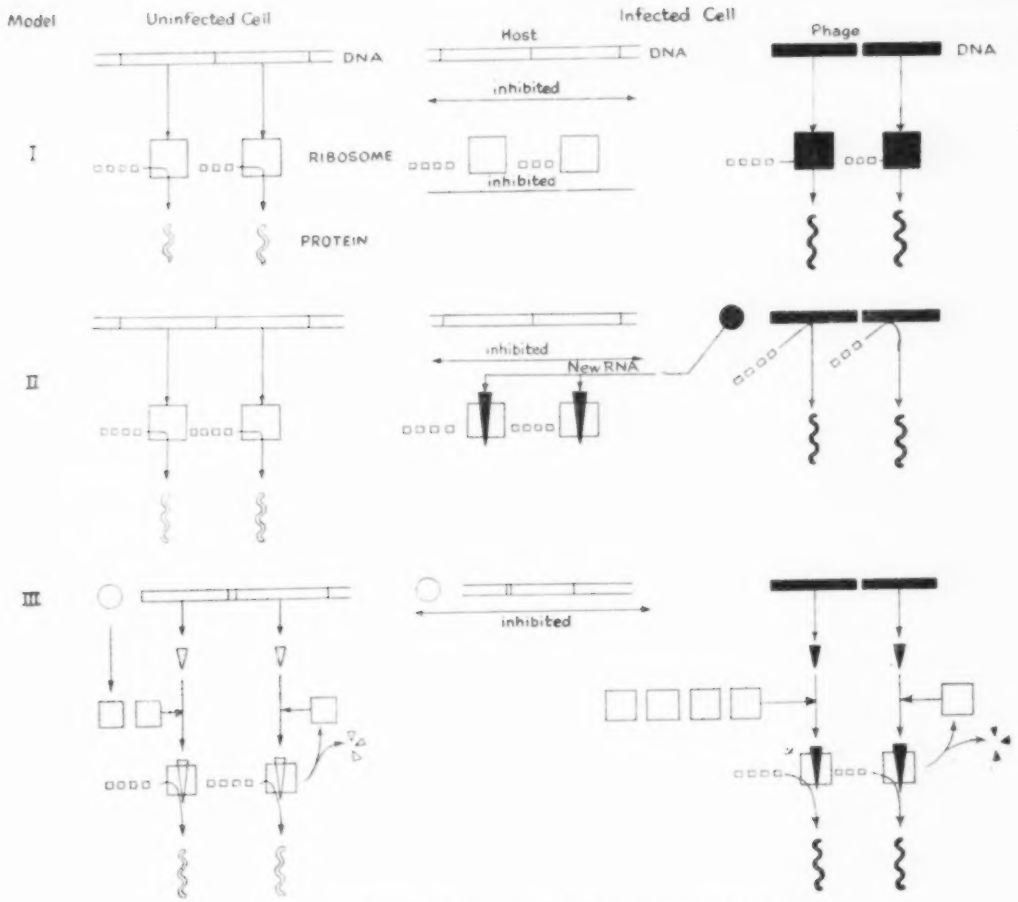

Fig. 1. Three models of information transfer in phage-infected cells

switched from bacterial to phage control and proceeds without the concomitant synthesis of stable RNA. *A priori*, three types of hypothesis may be considered to account for the known facts of phage protein synthesis (Fig. 1). Model I is the classical model. After infection the bacterial machinery is switched off, and new ribosomes are then synthesized by the phage genes. The *ad hoc* hypothesis has to be added that these ribosomes are unstable, to account for the turnover of RNA after phage infection. This is, in fact, the model favoured by Nomura *et al.*[12]. Model II assumes that in the particular case of phage the proteins are assembled directly on the DNA; the new RNA is a special molecule which enters old ribosomes and destroys their capacity for protein synthesis. At the same time, synthesis of ribosomes is switched off. Model III implies that a special type of RNA molecule, or 'messenger RNA', exists which brings genetic information from genes to non-specialized ribosomes and that the consequences of phage infection are two-fold: (*a*) to switch off the synthesis of new ribosomes; (*b*) to substitute phage messenger RNA for bacterial messenger RNA. This substitution can occur quickly only if messenger RNA is unstable; the RNA made after phage infection does turn over and appears, therefore, as a good candidate for the messenger.

It is possible to distinguish experimentally between these three models in the following way: Bacteria are grown in heavy isotopes so that all cell constituents are uniformly labelled 'heavy'. They are infected with phage and transferred immediately to a medium containing light isotopes so that all constituents synthesized after infection are 'light'. The distribution of new RNA and new protein, labelled with radioactive isotopes, is then followed by density gradient centrifugation[13] of purified ribosomes.

Density gradient centrifugation was carried out in a preparative centrifuge, and the ribosomes were stabilized by including magnesium acetate (0·01–0·06 *M*) in the caesium chloride solution. Ribosomes show two bands, a heavier *A* band and a lighter *B* band, the relative proportions of which, for a given preparation, depend on the magnesium concentration used. The lower the magnesium concentration, the smaller the proportion of *B* band ribosomes and the larger the proportion of *A* band ribosomes.

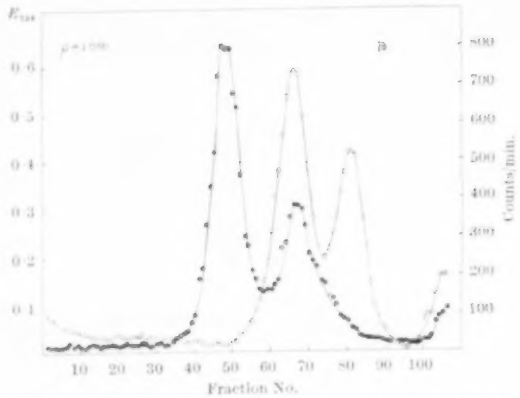

Fig. 2. Distribution of heavy and light ribosomes in a density gradient. *E. coli* B, grown in 5 ml. of a medium containing ^{15}N (99 per cent) and ^{13}C (60 per cent) algal hydrolysate and $^{32}PO_4$, were mixed with a fifty-fold excess of cells grown in nutrient broth, the ribosomes extracted by alumina grinding in the presence of 0·01 *M* Mg^{++} and purified by centrifugation. 1 mgm. of ribosomes was centrifuged in 3 ml. of cæsium chloride buffered to *p*H 7·2 with 0·1 *M tris* and containing 0·03 *M* magnesium acetate for 35 hr. at 37,000 r.p.m. in the SW39 rotor of the Spinco model *L* ultracentrifuge. After the run, a hole was pierced in the bottom of the tube and drops sequentially collected. Ultra-violet absorption at 254 mμ detects the excess of light ribosomes (○), ^{32}P counts detect the heavy ribosomes (●)

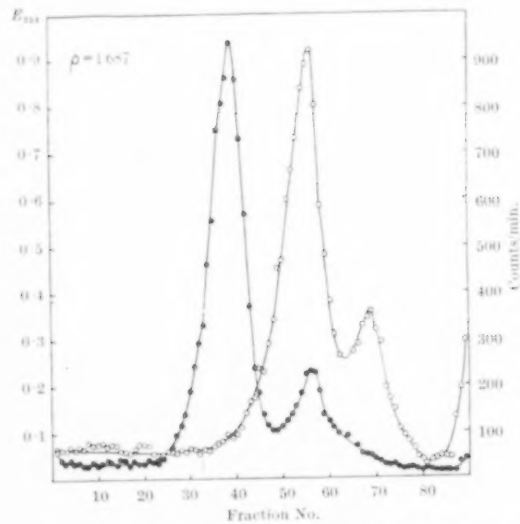

Fig. 3. Distribution of randomized heavy and light ribosomes in a density gradient. The mixture of $^{15}N^{32}P$ and $^{14}N^{31}P$ ribosomes was dialysed first for 18 hr. against 0·0005 *M* magnesium acetate in 0·01 *M* phosphate buffer *p*H 7·0, and then for 24 hr. against two changes of 0·01 *M* magnesium acetate in 0·001 *M tris* buffer *p*H 7·4. 1 mgm. of ribosomes was centrifuged for 38 hr. at 37,000 r.p.m. in cæsium chloride containing 0·03 *M* magnesium acetate. The drops were assayed for ultra-violet absorption (○) and ^{32}P content (●)

In order to show that there is no aggregation of ribosomes during preparation and density gradient centrifugation an experiment was carried out on ribosomes extracted from a mixture of $^{15}N^{13}C$ and $^{14}N^{12}C$ bacteria. The results are shown in Fig. 2, from which it can be seen that ribosomes of different isotopic compositions band independently and that there are no intermediate classes. The same preparation was then dialysed against low magnesium to dissociate the ribosomes into their 50 *S* and 30 *S* components and then against high magnesium to re-associate the sub-units[14]. This should have resulted in distributing heavy 30 *S* and 50 *S* sub-units into mixed 70 *S* and 100 *S* ribosomes. Surprisingly enough, density gradient centrifugation of this preparation (Fig. 3) yields the same bands as found in the original ribosomes except for a decrease in the proportion of the *B* bands. This means that both bands contain units which do not undergo reversible association and dissociation and that the mixed 70 *S* ribosomes prepared by dialysis separate

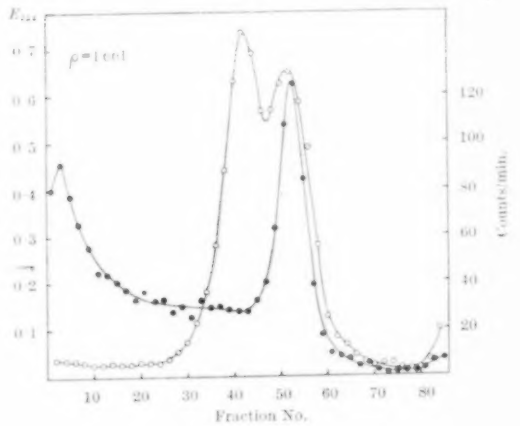

Fig. 4

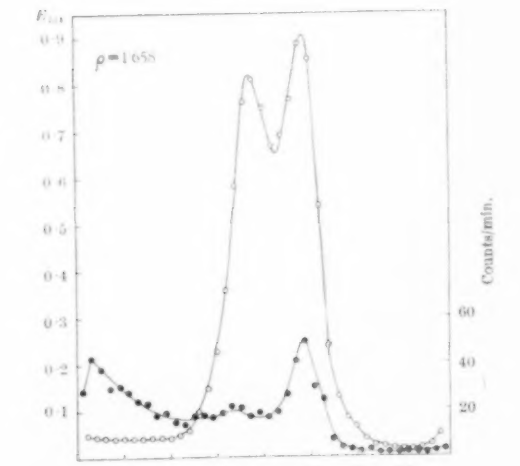

Fig. 5

Figs. 4 and 5. Distribution and turnover of RNA formed after phage infection. A 600 ml. culture of *E. coli B6* (mutant requiring arginine and uracil) was infected with *T4D* (multiplicity 30) and fed ^{14}C-uracil (10 mc./m*M*) from third to fifth min. after infection. One half of the culture was removed and ribosomes prepared (Fig. 4). The other half received a two hundred-fold excess of ^{12}C-uridine for a further 16 min. and ribosomes prepared (Fig. 5). In both experiments approximately 3 mgm. of purified ribosomes were centrifuged for 42 hr. at 37,000 r.p.m. in cæsium chloride containing 0·05 *M* magnesium acetate. Alternate drops were collected in *tris*-magnesium buffer for ultra-violet absorption (○) and on to 0·5 ml. of frozen 5 per cent trichloroacetic acid. These tubes were thawed, 1 mgm. of serum albumin added, and the precipitates separated and washed by filtration on membrane filters for assay of radioactivity (●)

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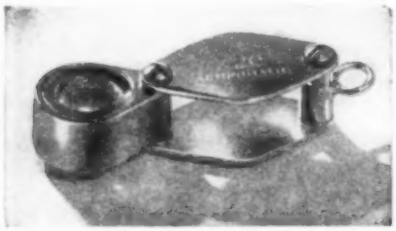

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into their components in the density gradient. Other experiments to be reported elsewhere suggest that the *A* band is composed of free 50 *S* and 30 *S* ribosomes and that the *B* band contains undissociated 70 *S* particles.

The bulk of the RNA synthesized after infection is found in the ribosome fraction, provided that the extraction is carried out in 0·01 *M* magnesium ions[12]. We have confirmed this finding and have studied the distribution of the new RNA among the ribosomal units found in the density gradient. Fig. 4 shows that this RNA, labelled with ^{14}C-uracil, bands in the same position as *B* band ribosomes. There is no peak corresponding to the *A* band. In addition, there is radioactivity at the bottom of the cell. This is free RNA as its density is greater than 1·8, and, moreover, it must have a reasonably high molecular weight to have sedimented in the gradient. Lowering of the magnesium concentration in the gradient, or dialysing the particles against low magnesium, produces a decrease of the *B* band and an increase of the *A* band. At the same time, the radioactive RNA leaves the *B* band to appear at the bottom of the gradient. This shows that the uracil has labelled a species of RNA distinct from that of the bulk of *B* band ribosomes, since the specific activity of the RNA at the bottom of the cell is much higher than that of the *B* band. Fig. 5 shows that this RNA turns over during phage growth. There is a decrease by a factor of four in the specific activity of the *B* band after 16 min. of growth in ^{12}C-uridine. Similar results have been obtained using $^{32}PO_4$ as a label.

These results do not distinguish between a messenger fraction and a small proportion of new ribosomes which are fragile in cæsium chloride and which are also metabolically unstable. In order to make the distinction, the experiment was carried out with an isotope transfer, in the following manner: Cells grown in a small volume of $^{15}N^{13}C$ medium were infected with *T* 4, transferred to $^{14}N^{12}C$ medium and fed $^{32}PO_4$ from the second to the seventh minute. They were mixed with a fifty-fold excess of cells grown and infected in $^{14}N^{12}C^{31}P$ medium. Fig. 6 shows that the RNA formed after infection in the heavy cells has a density greater than that of the *B* band of the carrier. Its peak corresponds exactly with the density of the *B* band of $^{15}N^{13}C$ ribosomes (Fig. 2) although it is skewed to lighter density, and its response to changing the magnesium concentration was that of a *B* band. There is no radioactive peak corresponding to the *B* band of the carrier: this means that no wholly new ribosomes are synthesized after phage infection. As already shown, the new RNA does not represent random labelling of *B* band ribosomes; therefore it con-

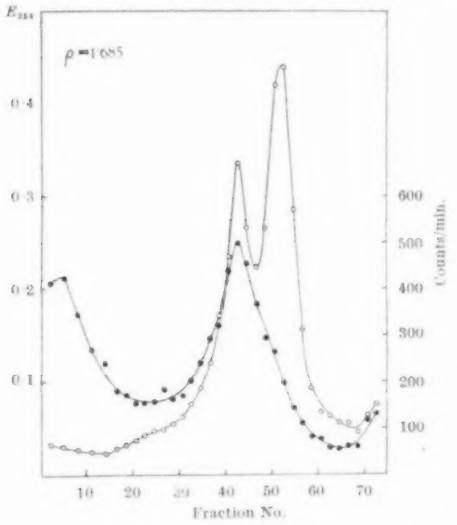

Fig. 6. RNA after isotope transfer in phage-infected cells. *E. coli* B grown in 10 ml. of ^{15}N (99 per cent) and ^{13}C (60 per cent) algal hydrolysate medium were starved in buffer, infected with *T* 4 and growth initiated by addition of glucose and dephosphorylated broth ($^{14}N^{12}C$). $^{32}PO_4$ was fed from the second to seventh min. after infection. The culture was mixed with a fifty-fold excess of *E. coli* B grown in nutrient broth, infected in buffer and then grown for 7 min. in dephosphorylated broth medium. 1 mgm. of purified ribosomes was centrifuged for 36 hr. at 37,000 r.p.m. in cæsium chloride containing 0·03 *M* magnesium acetate. Ultra-violet absorption (○) detects the $^{14}N^{12}C^{31}P$ carrier, radioactivity (●) detects the new RNA in the heavy cells transferred to light medium

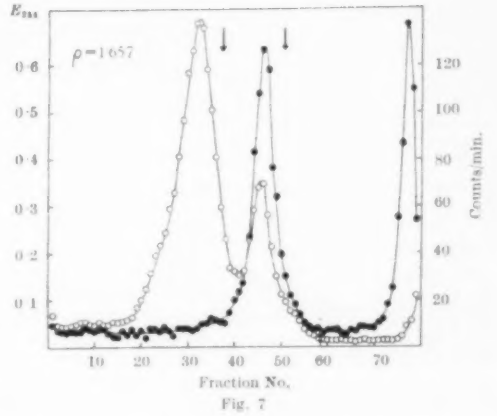

Fig. 7

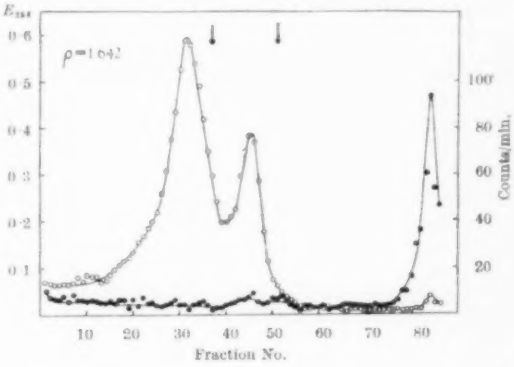

Fig. 8

Figs. 7 and 8. Distribution and turnover of newly synthesized protein in ribosomes of phage-infected cells after transfer. *E. coli* B grown in 600 ml. of a salt glucose medium containing $^{15}NH_4Cl$ (99 per cent) were starved in buffer, infected with *T* 4 and transferred to $^{14}NH_4Cl$ medium. $^{35}SO_4$ was fed for the first 2 min. of infection and one half of the culture removed (Fig. 7). The other half received an excess of $^{32}SO_4$ and ^{32}S-methionine and growth was continued for a further 8 min. (Fig. 8). 1 mgm. of purified ribosomes was centrifuged for 39 hr. at 37,000 r.p.m. in cæsium chloride containing 0·05 *M* magnesium acetate. Drops were assayed for ultra-violet absorption (○) and for radioactivity (●). The arrows mark the expected positions for the peaks of ^{15}N *A* and *B* bands. The radioactivity at the top of the gradient is contaminating protein

stitutes a fraction which is added to pre-existing ribosomes the bulk of the material of which has been assimilated before infection. This result conclusively eliminates model I.

To distinguish between models II and III an experiment was carried out to see whether pre-existing ribosomes participate in protein synthesis after phage infection. Cells were grown in ^{15}N medium, infected with phage, transferred to ^{14}N medium and fed $^{35}SO_4$ for the first 2 min. of phage growth. Fig. 7 shows that only the *B* band of pre-existing ribosomes becomes labelled with ^{35}S and there is no peak corresponding to a ^{14}N *B* band. All this label can be removed by growth in non-radioactive sulphate and methionine (Fig. 8). In this experiment, the incorporation of ^{35}S into the total extract was measured and the amount in the *B* band found to correspond to 10 sec. of protein synthesis. This is probably an over-estimate since it is unlikely that pool equilibration was attained instantaneously. This value corresponds quite closely with the ribosome passage time of 5–7 sec. for the nascent protein in uninfected cells[15]. In addition, electrophoresis of chymotrypsin digests of *B* band ribosomes shows that the radioactivity is already contained in a variety of peptides. It would therefore appear that most, if not all, protein synthesis in the infected cell occurs in ribosomes. The experiment also shows that pre-existing ribosomes are used for synthesis and that no new ribosomes containing stable sulphur-35 are synthesized. This result effectively eliminates model II.

We may summarize our findings as follows: (1) After phage infection no new ribosomes can be detected. (2) A new RNA with a relatively rapid turnover is synthesized after phage infection. This RNA, which has a base composition corresponding to that of the phage DNA, is added to pre-existing ribosomes, from which it can be detached in a cæsium chloride gradient by lowering the magnesium concentration. (3) Most, and perhaps all, protein synthesis in the infected cell occurs in pre-existing ribosomes.

These conclusions are compatible only with model III (Fig. 1), which implies that protein synthesis occurs by a similar mechanism in uninfected cells. This, indeed, appears to be the case: exposure of uninfected cells to a 10-sec. pulse of $^{32}PO_4$ results in labelling of the RNA in the *B* band and not in the *A* band of ribosomes, and this RNA can be detached from the ribosomes by lowering the concentration of magnesium ions. Similarly the nascent protein can be labelled by a short pulse of $^{35}SO_4$; it is located in the *B* band and most of the label is removed by growth in non-radioactive sulphate. In contrast to what was observed in infected cells, residual stable radioactivity is found in both bands, reflecting the synthesis of new ribosomes.

In order to act as an intermediate carrier of information from genes to ribosomes, the messenger has to fulfil certain prerequisites of size, turnover and nucleotide composition. In the accompanying article, Gros *et al.*[16] have analysed the distribution of pulse-labelled RNA in sucrose gradients. They have shown that in uninfected cells there is an RNA fraction which has a rapid turnover and which can become attached reversibly to ribosomes depending on the magnesium concentration. The *T2* phage-specific RNA shows the same behaviour and both are physically similar, with sedimentation constants of 14–16 *S*. We have carried out similar experiments[17] independently and our results confirm their findings. These suggest that, although the messenger RNA is a minor fraction of the total RNA (not more than 4 per cent), it is not uniformly distributed over all ribosomes, and may be large enough to code for long polypeptide chains. When ribosomes, from phage-infected cells labelled with $^{32}PO_4$ for five min., are separated by centrifugation in a sucrose density gradient[18] containing 0·01 *M* Mg^{++}, most of the messenger is found in 70 and 100 *S* ribosomes, contrary to previous reports[12]. When the magnesium concentration is lowered, the radioactivity is found in three peaks of roughly equal amount: (1) corresponding to a small residual number of 70 *S* ribosomes; (2) corresponding to 30 *S* ribosomes; (3) a peak of very high specific activity at 12 *S*. Separation of the RNA extracted from such ribosomes with detergent shows all the counts to be located in a peak at 12 *S*, skewed towards the heavier side. These results suggest that the messenger is heterogeneous in size and may have a minimum molecular weight of about $\frac{1}{4}$ to $\frac{1}{2}$ million. Similar results have been obtained in uninfected cells[17].

The undissociable 70 *S* ribosomes are enriched for messenger RNA over the total ribosomes of this type and it has been shown that they are also enriched for the nascent protein[17]. These ribosomes have been called "active 70 *S*" ribosomes, by Tissières *et al.*[19], and they appear to be the only ribosomes which preserve the ability to synthesize protein *in vitro*. This leads one to suspect that there is a series of successive events involved in protein synthesis, and that at any time we investigate a temporal cross-section of the process.

The exact determination of the rate of turnover of the messenger RNA should give information about the process of protein synthesis. This might be stoichiometric, in the sense that each messenger molecule functions only once in information transfer before it is destroyed. Its rate of turnover should then be the same as that of the nascent protein; but experiments to test this idea have been limited by difficulties in pool equilibration with nucleotide precursors.

It is a prediction of the hypothesis that the messenger RNA should be a simple copy of the gene, and its nucleotide composition should therefore correspond to that of the DNA. This appears to be the case in phage-infected cells[11,20], and recently Yeas and Vincent[21] have found a rapidly labelled RNA fraction with this property in yeast cells. If this turns out to be universally true, interesting implications for coding mechanisms will be raised.

One last point deserves emphasis. Although the details of the mechanism of information transfer by messenger are not clear, the experiments with phage-infected cells show unequivocally that information for protein synthesis cannot be encoded in the chemical sequence of the ribosomal RNA. Ribosomes are non-specialized structures which synthesize, at a given time, the protein dictated by the messenger they happen to contain. The function of the ribosomal RNA in this process is unknown and there are also no restrictions on its origin in the cell; it may be synthesized by nuclear genes or by enzymes or it may be endowed with self-replicating ability.

This work was initiated while two of us (S. B. and F. J.) were guest investigators in the Division of

Biology, California Institute of Technology, Pasadena, during June 1960. We would like to thank Profs. G. W Beadle and M. Delbruck for their kind hospitality and financial support.

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UNSTABLE RIBONUCLEIC ACID REVEALED BY PULSE LABELLING OF *ESCHERICHIA COLI*

By Drs. FRANCOIS GROS and H. HIATT

The Institut Pasteur, Paris

Dr. WALTER GILBERT

Departments of Physics, Harvard University

AND

Dr. C. G. KURLAND, R. W. RISEBROUGH and Dr. J. D. WATSON

The Biological Laboratories, Harvard University

WHEN *Escherichia coli* cells are infected with *T* even bacteriophage particles, synthesis of host proteins stops[1], and much if not all new protein synthesis is phage specific[2]. This system thus provides an ideal model for observing the synthesis of new proteins following the introduction of specific DNA. In particular, we should expect the appearance of phage-specific RNA, since it is generally assumed that DNA is not a direct template for protein synthesis but that its genetic information is transmitted to a specific sequence of bases in RNA. It was thus considered paradoxical when it was first noticed[3] that, following infection by the *T* even phages, net RNA synthesis stops even though protein synthesis continues at the rate of the uninfected bacterium. This could mean that DNA sometimes serves as a direct template for protein synthesis. Alternatively, net RNA synthesis may not be necessary so long as there exists the synthesis of a genetically specific RNA that turns over rapidly. This possibility was first suggested by experiments of Hershey[4], who, in 1953, reported that *T2* infected cells contain a metabolically active RNA fraction comprising about 1 per cent of the total RNA. Several years later, Volkin and Astrachan[5] reported that this metabolic RNA possessed base ratios similar, if not identical (considering uracil formally equivalent to thymine), to those of the infecting *T2* DNA. By 1958 they[6] extended their observation to *T7* infected cells, where again the RNA synthesized after phage infection had base ratios similar to those of the phage DNA.

During these years, evidence[7] accumulated that the sites of much, if not all, protein synthesis are the ribosomal particles, and it was thought most likely that ribosomal RNA was genetically specific, with each ribosome possessing a base sequence which coded for a specific amino-acid sequence (one ribosome–one protein hypothesis). Direct verification of this hypothesis was lacking, and its proponents[8] were troubled by the fact that, except for phage-specific RNA, it was impossible to find any correlation within a given organism between the base ratios of DNA and RNA. Moreover, there was no evidence that phage-specific RNA was ribosomal RNA.

Nomura, Hall and Spiegelman[9] have recently discovered that following *T2* infection there is no synthesis of typical (see below) ribosomal RNA and that the phage-specific RNA sediments at a slower rate (8*s*) than ribosomal RNA (16*s* and 23*s*). The genetic information for the synthesis of phage-specific proteins does not reside in the usual ribosomal RNA. Instead, if we assume that the synthesis of phage-specific proteins also occurs on ribosomes, then the phage-specific RNA might be viewed as a 'messenger' (to use the terminology of Monod and Jacob[10]) which carries the genetic information to the ribosomes. Furthermore, unless we postulate that there exist two different mechanisms for protein synthesis, there should also exist within uninfected normal cells RNA molecules physically similar to the phage-specific RNA and having base ratios similar to its specific DNA.

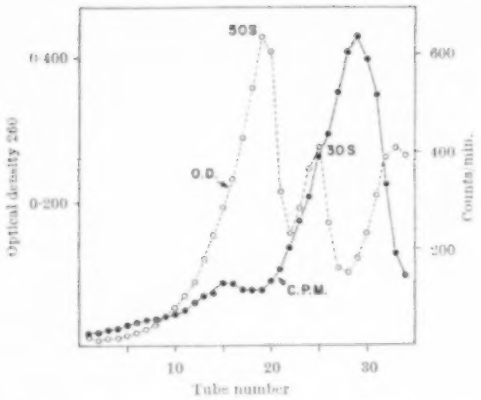

Fig. 1. Sedimentation of pulse-labelled 10^{-4} Mg^{2+} extract. 1 ml. of a crude extract labelled by a 20-sec. pulse of ^{14}C-uracil and made in 10^{-4} *M* Mg^{2+} and 5×10^{-4} *tris* buffer (*p*H 7·4) was carefully layered on top of a 24-ml. sucrose gradient (5–20 per cent) in a Spinco *SW*25 swinging-bucket tube. It was spun 2 hr. 45 min. at 25,000 r.p.m. at 4° C. The tube bottom was then punctured and 10 drop samples taken to measure 2600 Å. absorption (2 drops) and the radioactivity in 5 per cent trichloracetic acid precipitates (8 drops). All points were counted to at least 1,000 counts

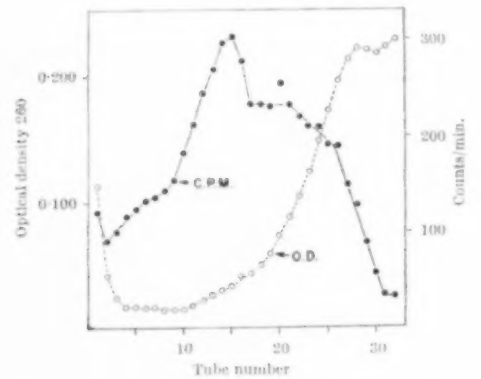

Fig. 2. Sedimentation of pulse-labelled 10^{-4} Mg^{2+} extract, long run. An extract as in Fig. 1 run for 11 hr. at 25,000 r.p.m., 15° C., on a sucrose gradient. Drop collection and radioactivity measurements were made as described in Fig. 1

Here we present evidence that RNA molecules physically similar to phage-specific RNA exist in normal *E. coli* cells and that, under suitable ionic conditions, they are associated with ribosomal particles.

Experimental plan. Most (80–85 per cent) RNA in actively growing *Escherichia coli* cells is found in ribosomal particles composed of 64 per cent RNA and 36 per cent protein[11]. There are two sizes[12] of ribosomal RNA, 16*s* (molecular weight = $5{\cdot}5 \times 10^5$) and 23*s* (molecular weight = $1{\cdot}1 \times 10^6$). The 16*s* RNA is derived from both 30*s* and 50*s* ribosomes, while 23*s* RNA is only found in 50*s* ribosomes. The other principal (10–15 per cent) form of RNA in *E. coli* is soluble RNA (now more appropriately called transfer RNA), which functions in the movement of activated amino-acids to the ribosomes. At least 20 (one for each amino-acid) different transfer RNA molecules exist[13], all of which have molecular weights about 25,000 and sedimentation constants of 4*s*.

Collectively, ribosomal and transfer RNA comprise at least 95 per cent of *E. coli* RNA. Thus messenger RNA, if present, can amount to at most only several per cent of the total RNA. Now if the messenger were stable, only a corresponding fraction of newly synthesized RNA could be messenger RNA; the great majority of new RNA being the metabolically stable ribosomal and transfer RNA's. If, however, messenger RNA is turning over (as is suggested by the original Hershey experiments) then a much larger fraction of newly made RNA must be messenger. For example, if the messenger functions only once for the synthesis of a single protein molecule, its lifetime might be only several seconds. In this event, if we look at the RNA synthesis occurring during a very short interval, then most of the newly synthesized RNA would be messenger even though this fraction may comprise only 1 per cent of the total RNA.

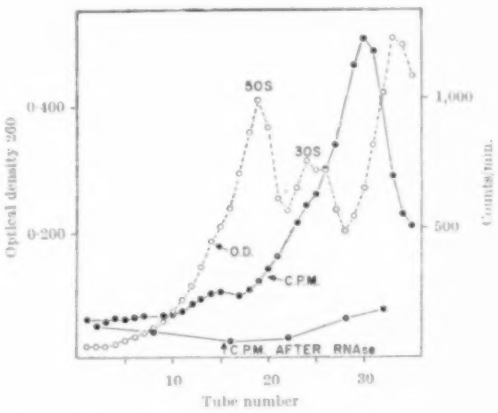

Fig. 3. Sedimentation of *T*2-infected 10^{-4} Mg^{2+} extract. An extract in 10^{-4} Mg^{2+} of cells infected with *T*2 at a multiplicity of 20 and labelled with phosphorus-32 between the second and fifth minutes after infection. Run as in Fig. 1. Several samples were treated with ribonuclease (10 γ/c.c.) to determine the background of label not in RNA

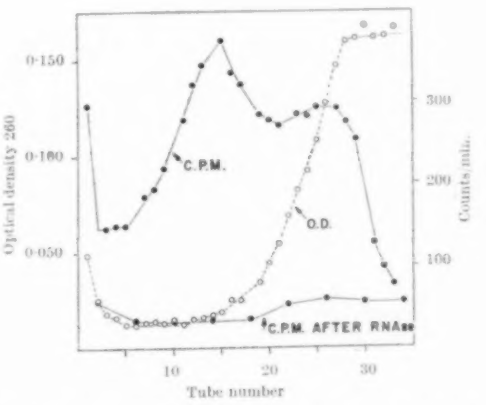

Fig. 4. Sedimentation of *T*2-infected 10^{-4} Mg^{2+} extract, long run. An extract as in Fig. 3 run for 11 hr. at 25,000 r.p.m. 20° C. Measured as in Fig. 1

We have therefore exposed *E. coli* cells to short pulses (10–20 sec. at 25° C. where the time of generation is about 90 min.) of radioactive RNA precursors (^{32}P or ^{14}C-uracil), rapidly chilled the cells with crushed ice and *M*/100 azide, prepared cell-free extracts by alumina grinding, added deoxyribonuclease at 5γ/ml., and examined the newly made RNA using the sucrose-gradient centrifugation technique[14]. In some experiments ^{14}C-uracil was given to cells of a pyrimidine-requiring mutant (*B*148) which was briefly starved (30 min. at 25° C.) for uracil. No difference has been seen between the properties of RNA labelled in these two ways. Starved cells incorporate more radioactivity, and they were used in most of the experiments reported below.

Results. Radioactive uracil (or phosphorus-32) is incorporated in RNA within several seconds after addition of the isotope. The RNA labelled by 10–20 sec. pulses is stable at 4° C. in cell-free extracts where ribonuclease and polynucleotide phosphorylase are not active. Traces (1γ/ml.) of ribonuclease degrade it under conditions where RNA in ribosomes is untouched. This suggests that it exists less protected than bound ribosomal RNA. Similarly, addition of phosphate permits the polynucleotide phosphorylase[15] in the cell extract to degrade preferentially pulse-labelled RNA. Our experiments thus as a matter of routine avoid the use of phosphate buffers.

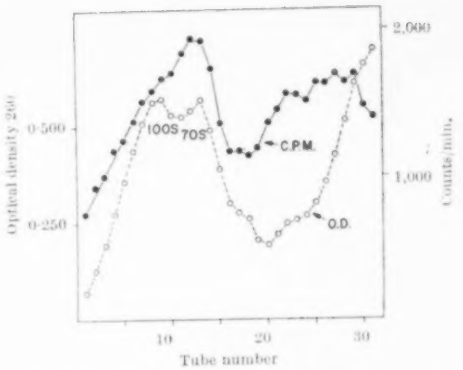

Fig. 7. Sedimentation of *T*2-infected 10^{-4} Mg^{2+} extract. An extract in 10^{-4} *M* Mg^{2+} of cells infected with *T*2 and labelled with phosphorus-32 from the second to the fifth minutes after infection. Run on a sucrose gradient in 10^{-3} *M* Mg^{2+} for 2 hr. 45 min. at 25,000 r.p.m. Drop collection and radioactivity measurements as for Fig. 1

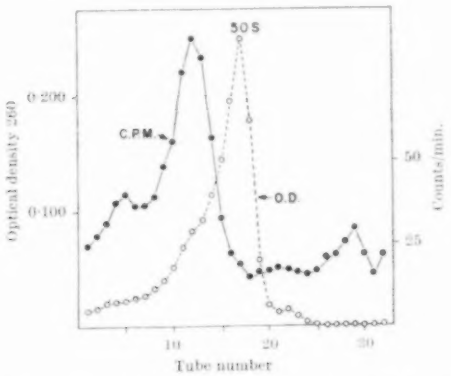

Fig. 5. Pulse-labelled active 70*s* ribosomes in 10^{-4} Mg^{2+}. An extract of cells labelled with a 20-sec. pulse of ^{14}C-uracil, made in 10^{-4} *M* Mg^{2+}, was purified by three 1 hr. centrifugations at 40,000 r.p.m. in 10^{-4} *M* Mg^{2+}, 5×10^{-3} *M tris* (*p*H 7·4). The resuspended pellet was run as for Fig. 1

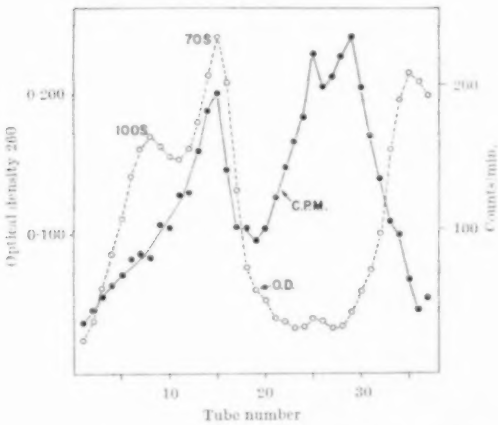

Fig. 6. Sedimentation of pulse-labelled 10^{-4} Mg^{2+} extract in 10^{-3} *M* Mg^{2+}. Magnesium was added to the 10^{-4} Mg^{2+} extract of Fig. 1 to bring it to 10^{-3} *M* Mg^{2+}. The sample was then run on a sucrose gradient in 10^{-3} *M* Mg^{2+}, 5×10^{-3} *M tris*, for 2 hr. 45 min. The measurements were made as described for Fig. 1

Figs. 1 and 2 show how RNA labelled by a 20-sec. exposure of uninjected cells to ^{14}C-uracil sediments in a sucrose gradient. The cell-free extract contains 10^{-4} *M* magnesium ions in which 30*s* and 50*s* ribosomes predominate. The majority of the radioactivity is not associated with ribosomes but moves with a 14–16*s* peak. There is also RNA which moves more slowly, in addition to a faster forward fraction moving at 70*s*. The slower sedimenting fraction shows up more clearly in the longer centrifugation shown in Fig. 2. Here a sharp 14–16*s* component is seen together with material sedimenting at 4–8*s*. Figs. 3 and 4 illustrate experiments with extracts from *T*2 infected cells exposed to phosphorus-32 from 2 to 5 min. after infection. They are similar, if not identical, to Figs. 1 and 2. Both have a major 14–16*s* component, a slower trailing fraction, and about 10 per cent of the material moving at 70*s*. The 70*s* component can be purified by three 1-hr. centrifugations at 40,000 r.p.m. with 10^{-4} *M* magnesium ions to concentrate the faster-moving ribosomes of Fig. 1. Fig. 5 shows the purified ribosomes consisting largely of 50*s* particles together with a now visible 70*s* component. The radioactivity, however, sediments as 70*s* ribosomes.

Still more label is attached to 70*s* particles when extracts are made in 10^{-3} *M* magnesium ions or when additional magnesium ions are added to 10^{-4} *M* extracts. Figs. 6 and 7 again illustrate the parallel appearance of normal and phage-infected extracts. About 30 per cent of label sediments with the 70*s* and 100*s* ribosomes, with the specific activity of 70*s* ribosomes generally greater than that of 100*s* ribosomes. When slightly lower concentrations of magnesium ions are used to give similar amounts of 30*s*, 50*s* and 70*s* ribosomes, label is specifically associated only with 70*s* particles and no label is in 50*s* ribosomes. The radioactivity sedimenting about

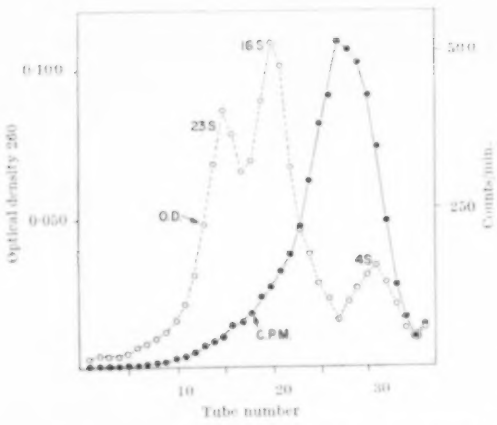

Fig. 8. Sedimentation of pulse-labelled RNA. An extract of cells labelled with a 20-sec. pulse of ^{14}C-uracil was treated with 0·5 per cent sodium lauryl sulphate ('Duponol') and then extracted three times with phenol. The RNA was then precipitated three times with alcohol and finally resuspended in 10^{-2} acetate buffer (pH 5·1). The RNA was run on a sucrose gradient, containing 10^{-2} acetate (pH 5·1) and M/10 sodium chloride, for 10 hr. at 25,000 r.p.m., 4° C. The drop collection and radioactivity measurements were made as described for Fig. 1

30s shows a very broad peak (20–40s) and it is probable that high concentrations of magnesium ions cause the 14–16s material either to aggregate or to assume a more compact shape.

Purified RNA, prepared by sodium lauryl sulphate ('Duponol') and phenol treatment of 20-sec. pulse extracts, sediments as shown in Fig. 8. Again the pattern is identical to that of labelled RNA from $T2$ infected cells (Fig. 9). Most newly synthesized RNA has a sedimentation constant about 8s; there is no appreciable synthesis of typical 16s or 23s ribosomal RNA. Since much new RNA in crude extracts moves at 14–16s, we considered the possibility that pulse-RNA is more sensitive than ribosomal RNA to degradation during the phenol extractions. Sodium lauryl sulphate (0·5 per cent) alone was therefore added to labelled crude extracts. This detergent treatment[12] both separates RNA from protein and completely inhibits action of the latent ribonuclease[13] release upon ribosome breakdown. When such

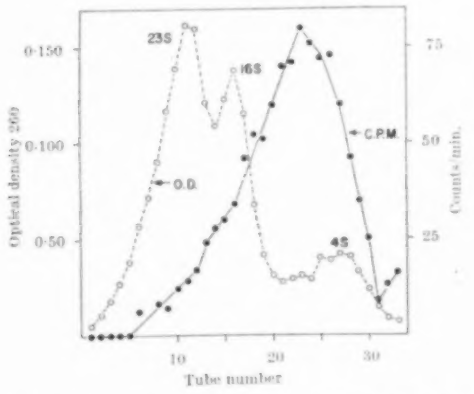

Fig. 9. Sedimentation of $T2$-specific RNA. RNA of cells infected with $T2$ and labelled with phosphorus-32 between the second and fifth minute, prepared and run as described for Fig. 8

treated extracts were run in sucrose gradients, the results were identical to those with phenol-prepared RNA.

The turnover of pulse-RNA is seen in chase experiments in which an excess of cold uracil is added after a 10-sec. ^{14}C-uracil pulse. After 15 min. at 37° C., all the label in crude extracts leaves the 14–16s component and is incorporated in the metabolically stable ribosomal and soluble RNA. In our kinetics experiments, the 16s RNA molecules always become labelled before appreciable labelling of 23s RNA molecules; likewise the 30s ribosomes become labelled before the 50s ribosomes. Since the base ratios of 16s and 23s chains are identical[17], it is likely that the 23s molecules form from two 16s chains.

Discussion. Our pulse experiments show that uninfected cells contain unstable RNA with sedimentation constants and attachment properties similar to those of $T2$-specific RNA. It is tempting to believe that these unstable molecules convey genetic information and are 'messenger' RNA. Complete homology with phage-specific RNA, however, will be demonstrated only if the base ratios of pulse-RNA are those of DNA. Unfortunately, in *E. coli*, the DNA base ratios do not differ greatly from ribosomal RNA, and as yet the base ratios of *E. coli* pulse-RNA are not precisely known. But we do have preliminary results (Hayes, D., and Gros, F., unpublished results) from *Staphylococcus aureus* which indicate an RNA component rapidly turning over and possessing DNA-like base ratios. Moreover, in yeast, there is also reported an unstable RNA resembling DNA in base composition[18]. We thus believe that our current measurements with *E. coli* will extend this fact and rule out the possibility that pulse-RNA is a precursor sub-unit from which ribosomal RNA is built up.

A messenger role for pulse-RNA fits nicely with its specific attachment to 70s ribosomes, the sites of protein synthesis[19]. Our experiments reveal two types of attachment. In one, the labelled RNA moves reversibly on or off 70s ribosomes depending on the concentration of magnesium ions. The second type of attachment binds pulse-RNA irreversibly to 'active' 70s ribosomes. These are 70s ribosomes which do not break apart with 10^{-4} M magnesium ions and which Tissières *et al.*[19] have shown to be the principal, if not sole, site of *in vitro* protein synthesis. They can be washed several times with 10^{-4} M magnesium ions without losing their ability to act in protein synthesis, and so they must have their genetic RNA firmly attached. It is reassuring that they can contain pulse-RNA. At least two stages, one reversible, the other not, thus exist in active 70s formation.

So far as we can tell, pulse-RNA, when associated with ribosomes, sediments at exactly 70s or 100s. Many experiments show that the label sediments neither faster nor slower than ordinary ribosomes. We find it hard to believe that the 14–16s RNA component which exists in 10^{-4} M magnesium ion extracts can combine with 70s or 100s ribosomes without altering their rate of sedimentation. Another possibility is that with 10^{-2} M magnesium ions, the 14–16s component assumes a more compact configuration similar to that of the RNA in a 30s particle. It could then associate with a free 50s ribosome to form a 70s ribosome which is transformed to an active 70s ribosome.

In conclusion, we state our findings: bacteria contain an RNA component turning over rapidly which

is physically distinct from ribosomal or soluble (transfer) RNA. This fraction behaves, in its range of sedimentation constants and its attachment to ribosomes in high magnesium ion concentrations, exactly as does the phage-specific RNA made after *T2* infection. Furthermore, it is associated with the active 70s ribosomes, the site of protein synthesis.

Our working hypothesis is that no fundamental difference exists between protein synthesis in phage-infected and uninfected bacteria. In both cases typical ribosomal RNA does not carry genetic information, but has another function, perhaps to provide a stable surface on which transfer RNA's can bring their specific amino-acids to the messenger RNA template.

These experiments were initiated when F. Gros was visiting the Biological Laboratories (May–August 1960). The pyrimidine-requiring strain *B148* was kindly provided by Dr. Martin Lubin. The financial support of the National Science Foundation and the National Institutes of Health is gratefully appreciated.

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DR. W. H. FITTON, F.R.S.

By JOAN M. EYLES

WILLIAM HENRY FITTON, physician and geologist, died in London on May 13, 1861, at the age of eighty-one. In an obituary notice, Sir Roderick Murchison, director-general of the Geological Survey, described him as "truly one of the British worthies who have raised modern geology to its present advanced position".

Fitton, of English descent, was born and brought up in Dublin and his proficiency in the classics enabled him to gain, in 1798, the Senior Scholarship of Trinity College. Even then he was a fossil collector, a hobby which brought him into the hands of the military on suspicion of being a rebel. In 1808, he went to Edinburgh to study medicine, and there attended the geological lectures of Robert Jameson, who was responsible for stimulating an interest in science and natural history in many young men at that time. Fitton graduated M.D. at Edinburgh in 1810, but continued to study medicine and chemistry in London until 1812. In that year he moved to Northampton, where he was in practice until 1820. Then marriage to a lady of considerable means enabled him to give up his practice and return to London, where his house soon became a regular meeting place for men of science and, in particular, geologists.

Fitton did not become a Fellow of the Geological Society until 1816, but his first geological paper, "On the Geological Structure of the Vicinity of Dublin", appeared in 1811 in the first volume of the Society's *Transactions*. In 1817 he contributed to the *Edinburgh Review* the first of a number of reviews, often lengthy, of geological works, which are noteworthy for the valuable information they contain on the history and progress of geology. These may still be consulted with profit by the historian of geology, for Fitton was widely read, had a clear analytical mind, and wrote with the facility of a classical scholar. Secretary of the Geological Society during 1822–24, he was elected president in 1827, and during his term of office introduced an important innovation, the publication of the *Proceedings* of the Society, of which the first number appeared in April 1827. In this publication the presidential addresses were published, Fitton's being the first to appear, and thus began that valuable annual series in which for many years the progress of geology was reviewed. Fitton was also instrumental in appointing, as curator and librarian, in 1829, William Lonsdale, a gifted palæontologist, who served the Society until 1842.

The geological work for which Fitton is best remembered, however, is his elucidation of the stratigraphy of the beds lying between the Kimmeridge Clay and the Chalk, that is, of the upper part of the Jurassic and the Lower Cretaceous. The succession comprises limestones, clays and sandstones, some marine and some fresh-water. During the course of a prolonged study of these beds in both the south-east of England and north-east France, he was able to establish the correct order of succession, a major advance in British stratigraphy. For these researches he received the Wollaston Medal of the Geological Society in 1852.

Fitton was elected Fellow of the Royal Society in 1815, and was also Fellow of the Linnean, Astronomical and Royal Geographical Societies.

OBITUARIES

Prof. Thomas Alan Stephenson, F.R.S.

Marine biologists in many lands will learn with sorrow of the death on April 3, at the age of sixty-three, of Prof. Thomas Alan Stephenson, for he had travelled widely and visited many ocean shores. Alan Stephenson's first interest was sea anemones, a group of animals which no doubt appealed to his artistic nature, and this culminated in the beautifully illustrated Ray Society monographs on British sea anemones which appeared in 1928 and 1935. He was on the staff of the Department of Zoology at University College, London, during 1922–30, and in 1923 made a short investigation for the States of Guernsey on the biology of the ormer, *Haliotis*, with the view of legislation to check the noticeable decrease in supply.

In 1928 he joined the Great Barrier Reef Expedition under C. M. Yonge and led the shore party concerned with the ecological investigations on the reefs. His knowledge of anemones made him most suitable for this expedition, and he naturally devoted much time to the corals themselves, making also valuable observations on growth. But his wider interest in shore ecology found full scope in a general study of reef ecology, on which one of his joint papers with his wife, G. Tandy and M. Spender, will have lasting value as evidence of the state of Low Isles in that year. It has already formed the basis of two later small-scale surveys.

In 1930, Stephenson was elected to the chair of zoology in the University of Cape Town. It was then that he developed his grand idea of the comparison of shore faunas on a world-wide scale. Our knowledge of the distribution of the intertidal fauna and flora had been restricted to detailed surveys at isolated points either in easy reach of marine laboratories or at places visited by expeditions. Stephenson was the first to organize a continuous survey along a great length of coast and its value lay in the fact that the same people saw the whole length of coast so that their observations were as comparable as possible.

This first great survey, over a period of ten years, extended from Port Nolloth on the west coast of Africa, round the Cape, to Durban on the east coast. It thus covered regions in the south Atlantic, the Southern and Indian Oceans, and extended through a range of climates as well as areas influenced by different ocean currents. The work was done with all his usual exactitude, and the correct identification of species was ensured so far as possible. The great series of papers produced by Stephenson and his colleagues will remain as a lasting memorial to a unique zoologist and as a reference point for all time. It was then that he discerned the underlying simplicity of the zonation patterns on the shore.

In 1940, he was appointed professor of zoology in the University of Wales at Aberystwyth, where he had been a student, and here he continued to inspire research on shore ecology. But every opportunity was still taken to continue his chief aim of seeing for himself as much of the coastline of the world as possible. With his wife Anne, who was always his constant and devoted collaborator, he explored the coasts of North America and of Bermuda. He was to have visited the Mediterranean this summer.

It was Alan's hope to write a book bringing together all his vast knowledge, and it is sad that he cannot now do this. But in fact a great deal has been published and is available to the world and the pioneering has been accomplished. The outburst of interest in marine science is ensuring that many more shores are being surveyed. The stimulus has been given and the standard set by Stephenson and his concept of three universal intertidal zones is being confirmed.

All Stephenson's work was characterized by its extreme finish, and everywhere could be seen his meticulous care for detail. But as well as being a first-class naturalist he was also an artist. His paintings of marine animals and plants are exquisite portrayals and their composition emphasizes his deep artistic appreciation and disclosure of pattern and rhythm, so beautifully exemplified in his little book on *Seashore Life and Pattern*.

The deepest sympathy of their many friends will go out to his wife in her bereavement.

F. S. Russell

Dr. Daphne Atkins

Dr. Daphne Atkins, a familiar figure to workers at the Plymouth Laboratory during the past 35 years, died on March 1. She was born on March 4, 1896, at Oxted, Surrey, the daughter of N. E. W. and Elisa Atkins. She was educated at the Kent County School for Girls, Tunbridge Wells, and from there went on to Bedford College, London, where she took her degree, in zoology and geology, in the early nineteen twenties.

Her first visit to Plymouth was probably in 1922 when I well remember her as a fellow student at the Easter class. After graduation she was soon to return to Plymouth where, supported initially by a series of research grants, she continued to work for much of the rest of her life. Her only notable period of absence was during and for some time after the War when she returned to Oxted to care for her aged parents.

She made major contributions to our knowledge of marine invertebrates. She observed the living animal, she studied its anatomy by dissection and in sections, in exact detail she followed the course of the ciliary currents concerned with respiration, feeding and cleansing in bivalve molluscs, polyzoans and brachiopods. She published her conclusions in admirably presented and most beautifully illustrated papers. These must have numbered at least forty and were largely published in the *Journal of the Marine Biological Association*, the *Quarterly Journal of Microscopical Science* and the *Proceedings of the Zoological Society of London*.

Her earliest research, suggested by J. H. Orton, on the moulting stages of pea-crabs (*Pinnotheres*) was published in 1927. While this led to further work on post-embryonic development, it had very different and far-reaching side issues. These included accounts of a fungal infection and of an entoniscid (Isopod) parasite on *Pinnotheres*, examples of contributions to parasitology which included descriptions of orthonectid, rhabdocoel and copepod parasites in bivalves.

Observing the effects of *Pinnotheres* on the gills of *Mytilus*, she noted that the damage caused led to the appearance of secondary or supernumerary food grooves with an associated reversal of beat in the adjacent frontal cilia. This led to further work on

abnormal gills and then on regeneration. These are very valuable papers containing information which could very suitably form the groundwork of further important studies. In the case of their author they led to a series of eight papers "On the Ciliary Mechanisms and Interrelationships of Lamellibranchs" published between 1936 and 1943. In the course of some 400 pages she ranged widely throughout the bivalves describing and figuring in the most precise and beautifully illustrated detail the course of ciliary currents and the different types of cilia, with such matters as cuticular fusion and the nature of ctenidial musculature. These papers are an abiding source of knowledge, one to which I am glad to pay my testimony of gratitude and admiration. They put all workers on the bivalve Mollusca, now and in the future, in her debt.

Another source of interest, again initially stimulated by J. H. Orton, was in the Polyzoa. Early in her research career she published an account of their ciliary feeding mechanisms followed by a detailed systematic and anatomical account of the Loxosomatidea of the Plymouth area. Later she was to describe the structure and feeding mechanisms of cyphonautes larvæ. Her latest interest, made possible by the more extensive collecting range of the post-war research vessel, *Sarsia*, was in Brachiopoda with which her last seven papers, dealing with development of the lophophore, with feeding mechanisms and with systematics, are concerned.

Although of an extremely retiring disposition, Daphne Atkins was far from being devoid of personality. Within the field of her research interests she held most decided views, supported as they were by the results of years of the most careful observation. She has left a magnificant monument in her long series of published papers. As the Plymouth Laboratory made her work possible, this itself is a justification for the existence of that Laboratory.

C. M. YONGE

NEWS and VIEWS

The First American Manned Ballistic Rocket Flight

ON May 5, at 14.34 hr. U.T., a *Redstone* rocket was launched from Cape Canaveral in Florida, carrying Commander Alan Shepard, of the U.S. Navy, in a space capsule. This first United States attempt at manned ballistic rocket flight was entirely successful. The capsule was accelerated to a maximum speed of 5,100 miles per hour, reached a height of 115 miles and travelled 302 miles down-range over the Atlantic. As it re-entered the atmosphere the capsule's heat-shield was seen from the waiting ships to be glowing red. Commander Shepard seems to have suffered no ill-effects as a result of his 15-min. flight, during which he was subjected to a deceleration of 11 *g*. He was in radio communication with the ground throughout, and was able to climb out of the capsule before a helicopter arrived to pick him up. Though this rocket firing was only a first step towards orbital flight, Commander Shepard was able to test orbital techniques by controlling the orientation of the capsule under zero-gravity conditions.

New Carbon Isotope, Carbon-16

A NEW carbon isotope of mass 16 has been discovered by physicists of the Atomic Weapons Research Establishment, Aldermaston, working in collaboration with scientists of the Clarendon Laboratory, Oxford. A carbon-14 target was bombarded with a beam of 6-MeV. tritons (ions of tritium) from the Aldermaston Van de Graaff accelerator. In some cases, the force of the collision was observed to be sufficiently great to split the triton into its constituents of one proton and two neutrons and the two neutrons were captured by the carbon-14 to form a new isotope, carbon-16. By making precise measurements with a large magnetic spectrograph on the emitted protons, the mass of the new isotope was found to be 16·014702 atomic mass units. It was anticipated that carbon-16 would suffer beta-decay like carbon-14 and carbon-15, but with the difference that the product of the decay would still have sufficient energy to emit a neutron. This occurred, and use was made of this property to measure the half-life, which was found to be 0·74 sec. The most abundant isotope of carbon is, of course, carbon-12. Isotopes of mass 13, 14 and 15 are known, of carbon-14 and -15 are radioactive, and the former is the basis for the carbon-14 dating of archæological specimens.

Ministry of Aviation : Mr. W. O. Broughton

MR. W. O. BROUGHTON has been promoted to deputy chief scientific officer and appointed to the post of director of air navigation and reconnaissance research and development in succession to Mr. C. J. Carter, who has joined the defence research staff in Washington. Mr. Broughton started his career in the Civil Service in 1935 at the Royal Aircraft Establishment, where he was engaged on development work connected with automatic pilots, the *Queen Bee* pilotless target aircraft and stabilized automatic bombsights. In 1942 he transferred to Headquarters, Ministry of Aircraft Production, to take charge of a section concerned with the development of automatic pilots, gyroscopes and stabilizing equipment. He returned to the Royal Aircraft Establishment in 1947 as head of the Controls Division of the Instrument and Air Photography Department, remaining in this post until 1954, when he was transferred to Headquarters, Ministry of Supply. In 1955 he was appointed assistant director, being responsible for administration of research and development connected with automatic flight control systems, automatic blind landing, flight instrument systems and aircraft fire control systems. In addition to these systems, Mr. Broughton now assumes responsibility for research and development on air photography, bombsights, flight simulators and non-radio aircraft navigation systems.

Physics at Birmingham: Prof. W. F. Vinen

DR. W. F. VINEN, who has been appointed to a chair of physics in the University of Birmingham at the early age of thirty-one, was educated at Watford Grammar School and Clare College, Cambridge, where he was elected to a research fellowship in 1955. In 1958, he was appointed a University demonstrator in physics and a Fellow of Pembroke College. In close collaboration with his contemporary, Dr. H. E.

Hall, recently appointed to a chair at Manchester (see *Nature*, March 18, p. 877). Dr. Vinen conducted an elegant series of studies on the hydrodynamics of superfluid helium, with particular reference to the development of turbulence and the frictional effects arising from the interactions of the thermally excited 'normal fluid' with the vorticity of the ground-state, the 'superfluid'. The work of these two collaborators, which must ultimately be ranked as a classic investigation, soundly establishing quantum hydrodynamics on an experimental basis, culminated on Dr. Vinen's side with a direct demonstration of the quantization of circulation in the superfluid. This beautiful experiment may well be regarded as a worthy conclusion to the work of more than twenty years in Cambridge on the properties of superfluid helium, which began equally strikingly with the discovery of the fountain effect. In the organization of a new department and the development of new fields of research, Dr. Vinen will have full scope for the inventiveness and versatility which have been such a notable characteristic of his work up to the present.

Director of the National Fishery Institute, Ecuador: Mr. F. Bourgois

Mr. François Bourgois has been appointed director of the National Fisheries Institute, Guayaguil (Ecuador), for a period of four years. Mr. Bourgois has been Food and Agriculture Organization fishery officer in Haiti since last July. Prior to that, he was fishery officer and the Organization's representative in Tunisia for three and a half years. While in Tunisia, Mr. Bourgois was concerned with finding and charting new fishing grounds, improving the sardine fisheries and extending Tunisian trawling farther offshore. Together with one of the Organization's master fishermen, he introduced the use of underwater lamps into Tunisian sardine fishing. Mr. Bourgois joined the French Fisheries Service in 1945 as a fisheries expert and administrator. He was previously fisheries adviser to the Viet Nam Government, and fisheries expert with the Supreme Commander for the Allied Powers. His first assignment for the Food and Agriculture Organization was in 1956 in Libya. The Ecuadorian National Fishery Institute is the second major U.N. Special Fund fisheries project, with the Food and Agriculture Organization as executing agency, to be established in Latin America. The project calls for a Government contribution of 663,040 dollars over a four-year period, and a special fund contribution of 633,800 dollars. The purpose of the Ecuadorian Institute is to help the Government foster the country's economic development by a substantial expansion of fishing and ancillary activities. Facilities and maintenance for the Institute's two research vessels, and personnel are to be provided by the Ecuadorian Navy. The Navy will also provide hydrographic and meteorological information on areas not studied by the Institute's vessels and will provide a ship for several months of whale-marking each year in the seas of Ecuador.

Space Research in Britain

Replying to a question regarding Great Britain's contribution to a space research programme, Mr. D. Freeth, Parliamentary Secretary for Science, said in the House of Commons on May 2 that Great Britain's programme comprises five main elements. There is the programme of scientific experiments using sounding rockets. Further experiments will be made in satellites to be launched by the United States, with which Britain is also collaborating in testing communications using satellites. Thirdly, Britain is participating actively in the preparatory commission set up to plan a European organization for co-operation in scientific research in space. Great Britain and France have also proposed to other European Governments a co-operative programme for the development of satellite launchers beginning with one based on *Blue Streak* to be fired from Woomera. Finally, Britain is doing much analysis of satellite tracking data, including the operation of one of the three World Data Centres which act as international clearing houses for information on space research. Future plans in Britain will be considerably affected by the results of the discussions now taking place with other countries regarding the third and fourth elements in this programme. Mr. Freeth added that Britain is in very close touch with both the Australian and Canadian Governments, and he strongly disagreed with the suggestion that discouragement of British participation in a space programme is leading to an exodus of the finest brains of the country to the United States and Canada.

In a written answer on May 2, Miss Pike, Assistant Postmaster-General, said that the Post Office is co-operating in a wide-ranging communications satellite research programme which covers orbits, altitudes and frequencies, and the design of the electronic equipment in the communication satellites themselves. The first requirement is a ground station for joint experiments with the Americans in satellite communications across the Atlantic.

In reply to questions on May 4, the Prime Minister said that the Minister for Science, in his speech to the Geological Society on April 26, was re-affirming his acceptance of the view of the Advisory Council on Scientific Policy that a full-scale space programme on a purely national basis, including the development of a large launcher, would not be justified solely on scientific grounds. The wider problem of developing a launcher for heavy satellites is under discussion in Europe, and any further statement of policy must await the outcome of discussions. He hoped that Britain would be able to get the joint scheme, because of the enormous sums involved if the matter were to be pressed forward in all its aspects. In other questions reference was made to the expenditure of only £5 million on scientific research by the National Coal Board, and to the cost of the Russian rocket programme for launching heavy satellites as about equal to the cost of the whole defence programme of Britain. Mr. Macmillan emphasized that it is a matter of determining the right priorities and in this matter the Government looks to the Advisory Council on Scientific Policy for guidance.

Industry, Science and the Chemical Engineer

On April 25, at the thirty-ninth annual general meeting of the Institution of Chemical Engineers, Mr. W. K. Hutchison, deputy chairman of the Gas Council, delivered the presidential address. He referred to the rapid growth in membership of the Institution. This occurred chiefly because of post-war expansion; chemical industry and chemical plant manufacturers all began to offer opportunities for employment to chemical engineers on a scale not contemplated before the War. The greatest demand is in the field of production, and, in particular, plant operation, where techniques of management are of special importance. The Institution is able to make one of its most useful contributions to the education

of engineers by providing the teaching institutions with detailed information on the type of problems encountered by the professional engineer. In time, each chemical engineering course offered by various institutions would develop its own style and character and the employer would remain free to go to whichever seemed best able to produce the type of man he wanted. The final test of the engineer would always be success or failure in carrying out the work entrusted to him. The Institution could do no more than prepare him for such a test.

Mr. Hutchison referred to the concept of 'unit operations' which, for thirty years, has been retained as a basis for instruction in chemical engineering. Behind all unit operations there are fundamental physical or chemical concepts, so related that, if used with the necessary degree of mathematical skill, they might make unnecessary much of the existing content of chemical engineering instruction. Mr. Hutchison also mentioned an interesting situation in continental Western Europe, where only small groups are willing to call themselves chemical engineers or to recognize the coherent set of principles within which a profession can be practised. Many of the functions claimed by chemical engineering are fulfilled by combined teams of chemists, physicists and engineers.

The attraction of chemical engineering to many young men is its obvious connexion with the realities of industrial processes. The attitude is that of the engineer rather than of the scientist; the laboratory and the lecture theatre are tolerated only as stepping-stones to the world outside. If there is a latent conflict between the two aspects of engineering, "the engineer directing the great powers in nature for the use and convenience of man", and the scientist intent in his laboratory on advancing the frontiers of knowledge, the Institution does provide a solution. It is both a corporate body of practising engineers and a meeting ground for scientists. The primary aim of the Institution is the advancement of science —by which is meant all knowledge—through the skill of practising engineers and scientists alike.

The Cotton, Silk and Man-Made Fibres Research Association

In May 1960, the working party that had been set up to consider the possible fusion of the British Cotton Industry Research Association and the British Rayon Research Association recommended that the two Associations should be merged into one research association as from July 1, 1961. It has now been decided that this will be known as the Cotton, Silk and Man-Made Fibres Research Association, and its main responsibilities will be to serve the processing and merchant converting sections of the industries. The officers of the new Association will be as follows: *Director*, Dr. D. W. Hill; *Deputy Director*, L. A. Wiseman; *Assistant Directors*, G. W. Lobb (to be responsible for research administration), L. H. C. Tippett (to be responsible for physical and mechanical research), and Dr. A. R. Urquhart (to be responsible for chemistry and chemical processing). A large part of the research programmes and other work previously undertaken by the original Associations is being continued by the new organization. The main source of the Research Association's industrial income will be a research levy forming part of the levy collected by the Cotton Board. The Association will also receive a grant from the Department of Scientific and Industrial Research. Plans are already well advanced for the provision of a new chemistry and finishing building at the Shirley Institute, Didsbury, Manchester, which, it is hoped, will be completed by the end of 1962 or early 1963.

Science Museum Exhibition of a Fourteenth-Century Astronomical Clock

THE Science Museum will be exhibiting during May 16–June 25 the first full astronomical clock to be produced in Britain for 250 years. The clock is an exact reproduction of one designed and constructed nearly 600 years ago by John Dondi, who was, at times, professor of logic, medicine and astronomy in Italy. During the years 1348–64, Dondi constructed his astronomical clock, which came to fame during the next two centuries, but afterwards it seems to have sunk into oblivion, and little was heard of it until the late G. H. Baillie, in the 1930's, translated the Latin manuscript which Dondi left behind giving a full description of just how he made the clock, with working drawings. The clock remained in Italy until 1585 and then went to Spain, where it was destroyed during the Peninsular Wars. Constructed within about fifty years of the invention of the mechanical clock, at a time when the standard clock was a clumsy forged-iron blacksmith's production, Dondi's clock presented an achievement of great refinement. Astronomical clocks showing the motions of the Sun, Moon and nodes began to be less rare in Europe during the late fifteenth and sixteenth centuries; but those embodying the motions of the five planets as well are rare at any period. The names of Immser and Baldewein came into prominence in Central Europe about 1550; in Western Europe little was done until Johann Klein of Prague and Aureliano and Cajetano of Vienna constructed their clocks in the last quarter of the eighteenth century. Later, clocks were built by Schwilgué in 1842 and Jensen in 1956, and in England there was only Samuel Watson in 1680 and Henry Bridges in 1730. It has fallen to Mr. H. Alan Lloyd to take up Baillie's work and complete it. After publishing a full précis of the translation in 1955, he decided to make a reconstruction of the clock. He was able to find collaborators in the managing director, Mr. Geoffrey Buggins, and in his co-director and chief craftsman, Mr. Peter Haward, of Thwaites and Reed, Ltd. Following the exhibition in London, the clock will be taken to the Smithsonian Institution, Washington.

X-ray Protection at High Voltages

THE National Committee on Radiation Protection and Measurement of the U.S. National Bureau of Standards has issued a new handbook with information about how to obtain medical protection from X-rays up to 3×10^6 V. (National Committee on Radiation Protection and Measurements. Report No. 26. Pp. viii + 52. (Washington, D.C., Government Printing Office, 1961.) 25 cents.) The handbook sets out standards of safety and contains data and recommendations relating to all persons involved, including the manufacturers of the apparatus and designers and builders of the rooms in which it is housed. Basic requirements for safety standards are essentially the same in this handbook as in its immediate predecessor (No. 60). Tables have been revised to conform with the new lower maximum permissible dose adopted by the Committee in 1957, and the differences in the maximum permissible doses allowed in controlled areas and their environs. Recom-

mendations have been changed to cover situations created by recent technical advances in machine design, and by the development of new radiological techniques. The underlying philosophy, however, remains as it was originally expressed by the Committee thirty years ago, namely, that unnecessary radiation exposure should always be avoided and all exposure kept to the minimum compatible with practical clinical requirements.

Field Studies in Scotland

Despite the attractiveness of the Study Centre at Garth, the Scottish Field Studies Association may have to discontinue its activities for lack of support. In contrast to the encouraging number of students who are attending Field Study Centres in England, the number of student days spent at Garth fell from 2,972 in 1959 to 2,539 in 1960. Of the 29 courses offered, 7 had to be cancelled for lack of support. The Association is appealing not only for financial help but also for increased support from Scottish and other local education authorities. In her foreword to the annual report, Dr. Margaret Stewart writes that "there is plenty of lip service to the importance of Field Studies in education curricula, but so long as it is left to the initiative of individual teachers the work of the Association cannot be developed on a firm and lasting basis" (Annual Report, 1960. The Importance of Field Studies in Modern Education. By Paul F. Holmes. Garth House as a Centre for Ecological Botany. By Fred N. and Brenda D. Haynes. Pp. 27+4 plates. (Glasgow: Scottish Field Studies Association, 1961.) 2*s*. 6*d*.). Information about the Association can be obtained from 179 West Regent Street, Glasgow 2.

Organization of Sandwich Courses

The National Advisory Council on Education for Industry and Commerce has started an inquiry on the organization of sandwich courses in technical colleges and other colleges of further education. A sub-committee under the chairmanship of Mr. E. L. Russell, chief education officer for Birmingham, has been set up "to examine the organization of sandwich courses with a view to using college staff, equipment and accommodation and industrial training facilities to the best advantage; and in particular to consider 'end-on' arrangements". The sub-committee is examining the various ways in which practical experience in industry is combined with academic study in technical college courses. These methods include the 'end-on' system by which two quite separate groups of students follow the same course, but use the college alternately, one group being in college while the other is in industry. Any organization or persons wishing to submit evidence to the sub-committee is asked to write to the Secretary, National Advisory Council on Education for Industry and Commerce, Ministry of Education, Curzon Street, London, W.1.

Zoological Society of London

During 1960 the Zoological Society of London was the recipient of three munificent bequests which will enable it to extend considerably its activities. The Trustees of the Nuffield Foundation granted the Society £100,000 to found an Institute of Comparative Medicine. Mr. Jack Cotton is providing £250,000 to help the rebuilding programme, and the Ford Foundation gave 550,000 dollars (nearly £200,000) to endow research fellowships in animal breeding. A major part of the new research activities will lie in the field of comparative medicine. The Institute of Comparative Medicine is to be associated with the name of Lord Nuffield. The grant is also intended to allow for the creation of two research fellowships in comparative medicine. The Ford grant makes it possible to undertake a systematic study of the reproductive habits and physiology of those animals in the Society's collections which breed readily in captivity. These studies will be supervised by a committee of well-known scientists distinguished for their contributions to knowledge of reproductive physiology. On the basis of the advice of this committee, the Council has already appointed the first three research Fellows. The Society is now planning new laboratories for their accommodation. Mr. Cotton's gift is the key which sets in motion the plan for reconstruction of the Regent's Park Gardens as devised by Sir Hugh Casson and Mr. Peter Shepheard in consultation with the Society's resident architect, Mr. F. A. Stengelhofen. Details of the bequests and other information about the many forward-looking activities of the Society are set out in its annual report for 1960 (Pp. 54+8 plates. London: Zoological Society of London, 1961). H.R.H. the Duke of Edinburgh has accepted the presidency in succession to Sir Landsborough Thomson.

The Ciba Foundation

The report for 1960 of the Ciba Foundation for the Promotion of International Co-operation in Medical and Chemical Research, which includes, as usual, details of symposia and their participants, of the study groups and research meetings, and lists of publications held and issued, records that the Foundation provided accommodation during the year for 825 scientists from 37 countries (Pp. 63. London: Ciba Foundation for the Promotion of International Co-operation in Medical and Chemical Research, 1961). Five three-day international conferences were held and membership of these symposia is now limited to about 30. Besides four one-day study groups, the Foundation arranged five evening discussion meetings, five research meetings, a guest symposium on "Aspects of Ciné-Indoscopy and Ciné-Radiology", seven sessions of scientific films, and a new series of dinner meetings known as the Dunant Coterie. Three such meetings were held in 1960 and the twelfth Ciba Foundation Lecture was delivered by Prof. J. Konorski, the fourth annual lecture on ageing by Dr. P. J. Thung, and the Louis Rapkine Lecture by Medicin-Lieutenant M. Flandet. Seven Anglo-French bursaries were awarded by the British selection committee for medical graduates to visit France, and two by the French selection committee for visits to London Medical Schools. Three volumes of the proceedings of symposia, one of a colloquium on endocrinology and two of study groups were published during the year.

Photon Rocket

Optical and mechanical phenomena to be encountered by a space traveller moving with a velocity approaching that of light are discussed by S. M. Rytov (*Priroda*, **4**, 64; 1960). The greatest danger to such a traveller will be encounters with relativistic cosmic dust and wind. A collision of the space ship with a cosmic particle one milligram in weight will spell complete destruction. In spite of such a warning G. G. Zolkin describes the construction of a purely hypothetical photon rocket (*Priroda*, **11**,

69; 1960), many kilometres in length and propelled by fuel, the energy of which is wholly transformed into luminous energy.

A New Elementary Particle

In a brief note Din Da-Tzao and E. N. Kladnitzkaya (*Priroda*, **8**, 17; 1960) give an account of the discovery in the Dubno laboratory of a new elementary particle—an antisigma minus hyperon. This discovery was made by a group of research workers under the leadership of V. I. Weksler and Van Gan-Chan. As shown by the particle track photograph this particle was produced through the collision of a pi-meson with the nucleus of a carbon atom.

French Scientific and Technical Books 1950–60

A collected catalogue of French scientific and technical books 1950–60, with an introduction by P. Piganiol, comprises a selection of books at present on sale and published between 1950 and 1960 (*Catalogue Collectif des livres Français de Sciences et Techniques*, 1950–1960. Pp. viii+242. Paris: Cercle de la Librairie, 1961). The books are classified by subject in five main sections under science, and eleven under technology, and alphabetically by title within the sub-sections. There are alphabetical subject and author indexes.

The Society for General Microbiology

The Society for General Microbiology held its annual general meeting on April 10, at the Royal Institution. For the forthcoming year the following officers were elected: *President*, Prof. R. Lovell; *Treasurer*, Prof. K. E. Cooper; *Editors*, Prof. B. C. J. G. Knight and Mr. A. F. B. Standfast; *Meetings Secretary*, Dr. B. W. Lacey; *General Secretary*, Dr. S. T. Cowan. The Marjory Stephenson Memorial Lecture, "The Development of Microbiology", was given by Prof. B. C. J. G. Knight, and a symposium on the Microbial Reaction to Environment was held on April 11 and 12. The subject of the Society's symposium in 1962 will be "Microbial Classification".

Electron Microscopy

The Electron Microscopy Group of the Institute of Physics and the Physical Society is holding its annual conference at the University of Nottingham during July 10–14. The conference will be concerned mainly with the physics of the electron microscope and its applications. Among the main topics to be discussed are: "Electron Microscopes and Electron Optics"; "Electron Microscopy of Thin Films"; "The Theory and Scope of Electron Diffraction Methods"; "Miscellaneous Techniques and Applications of Electron Microscopy". The conference will also include a symposium on "The Physics of Sectioning and the Embedding Process". Further information can be obtained from the Administration Assistant, Institute of Physics and the Physical Society, 47 Belgrave Square, London, S.W.1.

Seminars on Nuclear Science

The European Nuclear Energy Agency of the Organization for European Economic Co-operation has arranged with the Atomic Energy Research Establishment at Harwell and the Centre d'Etudes Nucléaires at Saclay for two international seminars on nuclear energy. Designed for the teaching staffs of universities and higher technical colleges, each seminar will accept about fifty applicants, who will have the opportunity to study the latest advances in nuclear science and techniques, to do certain practical work, and to visit nuclear installations. The Harwell seminar will take place during July 10–21, and that at Saclay, June 26–July 7. The costs of both seminars are being borne by the European Nuclear Energy Agency, and participants will only be required to pay travelling and living expenses. Further information can be obtained from the Organization for European Economic Co-operation, European Nuclear Energy Agency, 38 Boulevard Suchet, Paris 16e.

Announcements

Prof. K. E. Bullen, professor of applied mathematics in the University of Sydney, has been awarded the William Bowie Medal for Unselfish Co-operation in Research by the American Geophysical Union, in recognition of his contributions to knowledge of the interior of the Earth.

Dr. K. J. Irvine is to succeed Mr. W. E. Bardgett, at present research manager for research and development of the United Steel Companies, Ltd., when the latter retires on July 1. Dr. Irvine will take the title of metallurgical research manager.

The Institute of Fuel is organizing a second conference on "Waste Heat Recovery", which will be held in Bournemouth during May 15–19. Further information can be obtained from the Institute of Fuel, 18 Devonshire Street, Portland Place, London, W.1.

The theme of the 1961 convention of the British Institute of Radio Engineers is to be "Radio Techniques and Space Research". The Convention is being held in the University of Oxford during July 5–8. Further information can be obtained from the General Secretary, British Institution of Radio Engineers, 9 Bedford Square, London, W.C.1.

The Royal Meteorological Society is holding a two-day meeting at the University of Exeter during July 12–14. The meeting will include symposia on "Winds and Circulation in the Upper Atmosphere" and "Medium-scale Diffusion in the Atmosphere". Further information can be obtained from the Assistant Secretary, Royal Meteorological Society, 49 Cromwell Road, London, S.W.7.

The fifth international conference on "Ionization Phenomena in Gases", sponsored by the Association of the German Physical Societies (Plasma and Discharge Physics Group), will be held at Munich during August 28–September 1. Further information can be obtained from Prof. K. H. Hocker, Technische Hochschule Stuttgart, Herdweg 51, Stuttgart.

The 1961 biennial conference of the Oil and Colour Chemists' Association is to be held at Torquay during May 30–June 3. The general title of the conference will be "Physics in Surface Coatings". Further information can be obtained from the General Secretary, Oil and Colour Chemists' Association, Wax Chandlers' Hall, Gresham Street, London, E.C.2.

Errata. In the note entitled "The Geological Society of London", appearing on p. 492 of May 6 issue of *Nature*, the final sentence refers to newly elected members of the Society's Council. Furthermore, Dr. P. A. Sabine, one of the newly elected secretaries, is the chief petrographer, H.M. Geological Survey.

THE SCALE OF THE SOLAR SYSTEM

By THE STAFF, MILLSTONE RADAR OBSERVATORY, LINCOLN LABORATORY*,
MASSACHUSETTS INSTITUTE OF TECHNOLOGY,
WESTFORD, MASSACHUSETTS

THE close approach of Venus during April 1961 has permitted a re-determination of the scale of the solar system by means of radar distance measurements of this planet. Previous measurements of this kind have been made by Price *et al.*[1] and Evans and Taylor[2]. These earlier experiments led to a value for the solar parallax of $\pi = 8{\cdot}8022 \pm 0{\cdot}0001$ sec. of arc. This is not in agreement with new measurements made using the Millstone Radar Observatory of the Massachusetts Institute of Technology, Lincoln Laboratory, located at Westford, Massachusetts, in March and April 1961.

In all the experiments the transmitter is operated for a period somewhat longer than the two-way travel time. Following the transmission interval (usually about 5 min.) the received energy is integrated at different range-intervals for a duration corresponding to the echo time. In this way most possible sources of spurious echoes are removed. Allowance is made in the summation process for the gradual drifting of range with time. A display of the results of thirty such 'runs' for April 8 is shown in Fig. 1. Included in the processing is a calibration pulse, chosen to approximate the expected intensity of the

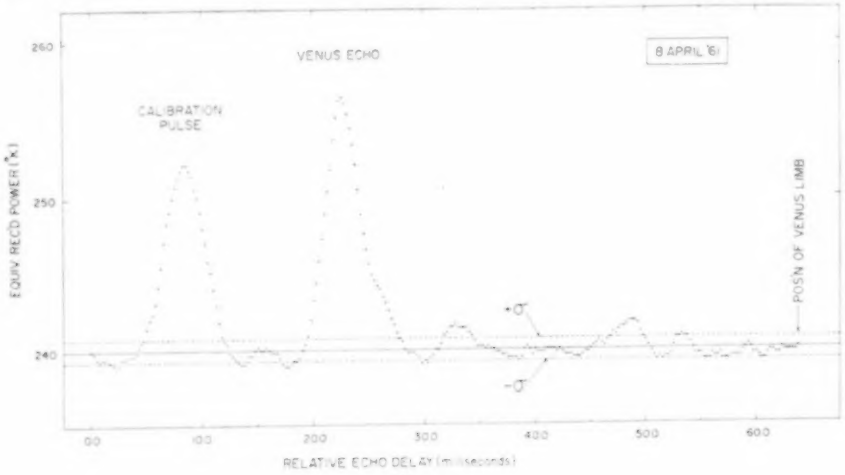

Fig. 1

This equipment operates with the following parameters: frequency, 440 Mc./s.; aerial gain, 37·5 db.; peak transmitter power, 2·5 MW.; receiver noise temperature, 240° K.; transmitter pulse-lengths, 0·5, 2, or 4 msec. The transmitter interpulse period has been varied at approximately daily intervals during the course of experiments which began on March 6. In this way the uncertainty in the total number of interpulse periods in the time for the pulse to travel the two-way distance to and from Venus can be removed. The value for the range of the planet has been converted to a value for the solar parallax by using an ephemeris computed in a manner described by Pettengill and Price[3]. The preliminary value obtained from twenty independent experiments conducted during the period March 6–April 12 is:

$$\pi = 8{\cdot}79450 \pm 0{\cdot}00008 \text{ sec. of arc}$$

which corresponds to a mean solar distance of 149,597,700 ± 1,500 km. The observations are being continued and it is expected that a final analysis of all the measurements will yield a value substantially more accurate than the new value reported here.

* Operated with support from the U.S. Air Force.

Venus echo. The Venus echo may be seen to rise some 23 standard deviations above the mean noise-level. The echo intensity corresponds to a received power-level of 163 db. below 1 mW.

From the intensity of the echo we conclude that Venus scatters about 12 per cent of the incident energy. This is not greatly different from the amount scattered by the Moon (7 per cent, ref. 4) and suggests that Venus has a solid surface.

A frequency analysis of the returned signal power discloses very little spectral broadening. Two 4-min. runs analysed to date show that essentially all the returned power is contained within a band-width of less than one cycle per sec. Until some measure of the angular scattering law is obtained it is impossible to determine the planetary rotation. It is interesting, to note, however, that if one assumes an angular distribution similar to the Moon, an apparent rotational period consistent with the synodic period (584 days) is obtained.

[1] Price, R., Green, P. E., Goblick, T. J., Kingston, R. H., Kraft, jun., L. G., Pettengill, G. H., Silver, R., and Smith, W. B., *Science*, **129**, 751 (1959).
[2] Evans, J. V., and Taylor, G. N., *Nature*, **184**, 1358 (1959).
[3] Pettengill, G. H., and Price, R., *Planetary and Space Science*, **5**, 70 (1961).
[4] Fricker, S. J., Ingalls, R. P., Mason, W. C., Stone, M. L., and Swift, D. W., *J. Research (Nat. Bur. Stand.)*, **64** D, 455 (1960).

FUNGICIDES IN AGRICULTURE AND HORTICULTURE

FROM time to time estimates are published of the enormous annual loss of crops throughout the world due to pests and diseases. A considerable proportion of these losses, equal to the produce of millions of acres of agricultural land, arises from fungal diseases, and in most cases the use of fungicides provides the most effective remedy. In recent years there has been good progress in the development of new fungicides; crop losses are still great, however, even in Britain, where potato blight, for example, still destroys thousands of tons of potatoes each year. Meetings concerned with research on fungicides do not occur frequently in Britain and considerable importance was therefore attached to the symposium entitled "Fungicides in Agriculture and Horticulture" organized by the Pesticides Group of the Society of Chemical Industry and held in London during March 20–21.

The programme was well balanced to include treatment of fundamental as well as practical aspects of the subject. The three papers delivered on the first morning under the general heading of "Current Usage and Recent Developments" and dealing with fruit, glasshouse crops and agriculture respectively were mainly practical. It is well known that organic fungicides have been used increasingly in crop protection in recent years, but it was interesting to hear from these three speakers that inorganic materials such as elemental sulphur, lime-sulphur and Bordeaux mixture still hold a very important position.

Dr. A. H. M. Kirby dealt briefly with soft and stone fruits and more fully with pome fruits. Scab has been the disease requiring most attention on apple and pear, but of all the chemicals used for its control none is without some defect. For example, captan, which gives excellent control, allows mildew to increase—this, incidentally, is now the most important disease for which a safe, cheap and adequate control is still required. The dangers of phytotoxicity and mammalian toxicity with some of the materials in current use were emphasized, and a particular point made was that the harmonizing of biological and chemical control of pests and diseases is worthy of greater attention in spray programmes.

In glasshouse crops, as W. H. Read pointed out, a system of monoculture is usually practised and the methods and costs of partial soil-sterilization to combat the soil-borne diseases encouraged by this system were given. The disadvantage with chemical soil sterilants is the long period required for the chemical or its breakdown products to disappear from the soil.

Since most glasshouse crops are susceptible to phytotoxicity and mechanical damage, there is usually no definite spray programme in glasshouses, the policy being to wait until disease needs to be controlled. The uses of sprays, aerosols, smokes and vaporization were discussed as methods of dispersing the chemicals employed to control a number of important diseases of glasshouse crops.

Unlike the previous speakers, Dr. F. C. Gayner was not restricted to considering the use of fungicides in Britain. Under the title "Fungicide Usage in World Agriculture" he gave the amount and value of sulphur, copper, mercury and organic fungicides used throughout the world during 1958. Although 12,000 tons of organic fungicides and 300 tons of mercury were used in that year, 275,000 tons of sulphur, used mostly as a dust, and 56,000 tons of copper were sold, the latter mainly as copper sulphate for the manufacture of Bordeaux mixture. Most of this interesting paper was concerned with the use of fungicides to control the eight major diseases on crops of world importance.

The second session of the symposium comprised three fundamental papers on "Theoretical Bases for Fungicides". The first of these, prepared by Dr. A. H. McIntosh, was read by Dr. C. Potter; but in the absence of the author it was inevitable that the discussion suffered some limitations. The results were given for the fungistatic action of nineteen chemicals, some of them well-known fungicides, against *Botrytis fabae* at temperatures between 10° and 25° C. and at intervals of up to 14 days. In general, an increase in temperature was shown to increase the toxicity and speed of action, whereas the apparent toxicity at any one temperature decreased as time passed. It emerged that there is little evidence for the generally held belief that spores are more resistant to fungicides at their optimum temperature for germination and also that the results may be misleading if spore germination tests are assessed before the ED_{50} has been allowed time to reach a steady value. An important point was made here concerning the loss of fungicides by sorption on the glassware used in spore-germination tests. This is particularly true of heavy-metal salts and, in one case cited, using mercuric chloride, some 70 per cent of the original amount was lost from solution in 24 hr.

Physical measurements and biological behaviour also formed an integral part of the succeeding paper by Dr. M. J. Janssen of Utrecht. Work on fungicides in the Netherlands has long been in the forefront of world research, and in recent years a series of papers dealing with the dithiocarbamate fungicides has been published by the team led by Prof. G. J. M. van der Kerk. Dr. Janssen's contribution was concerned with the mode of action of the dithiocarbamates, and he divided the work on this subject into three phases, dealing mainly with the most recent work relating to metal complex formation. He showed that the dithiocarbamates act in much the same way as 8-hydroxyquinoline, for which the importance of metal chelation has been demonstrated by Albert. A quantitative agreement exists between toxicity and concentration of the dithiocarbamate ion itself and its complexes with copper. Thus the different growth pattern in several fungi can be ascribed to their sensitivity not only to the dithiocarbamate ion in the medium but also to the complexes in which one or two dithiocarbamate ions are attached to the bivalent copper ion. Reference was also made in this paper to the current fashion of postulating metal-binding as responsible for the observed phenomena in other biological systems. In the discussion, there was general support for Dr. Janssen's opinion that the theory that chelation is of basic importance in relation to the activity of plant growth-regulating substances is unwarranted.

Dr. E. Somers pointed out that in terms of weight of chemical removed from solution by fungal spores, the fungicides in current use possess low toxicity compared with animal poisons, insecticides or even bactericides. He felt that the common method of determining uptake of fungicide by measuring the amount remaining in solution after fungal spores have been removed from it gives little information on the actual amount which has penetrated the spores. Using captan and a series of ring-substituted derivatives no correlation could be established between lipid solubility on one hand, and uptake or fungitoxicity, in terms of the ED_{50}, on the other. It was shown that thiol-groups within the cell are concerned in the action of captan ; furthermore, the chemical appears to be taken up only by living cells. This is quite different from the results obtained with radioactive copper, the uptake of which was found to be non-specific. Thus, any part of the fungal cell, whether living or dead, will apparently remove copper-64 from solution ; that which accumulates on the outside of the cell therefore probably plays no part in the fungicidal activity of this metal.

The first paper on the second day of the symposium, given by Prof. G. J. M. van der Kerk of Utrecht, was concerned with new developments in organic fungicides. An authority in this field, Prof. van der Kerk first described how fungicide research in Holland is co-ordinated between several institutions. He then went on to forecast that another general group of organic fungicides superior to the dithiocarbamates on grounds of activity, simplicity of molecular structure and cost is unlikely to be found. He considered it more likely that in the future specific materials will be developed to control specific diseases.

In the field of protectant fungicides, the organo-tin compounds have recently been introduced commercially as a result of research which started in 1950. Triphenyltin acetate is already marketed by Germany for use on a wide range of crops, but its phytotoxicity and mammalian toxicity require special attention. The triphenyltin sulphides, on the other hand, are much less toxic towards higher plants and animals but their antifungal spectra also show more specificity.

In the search for systemic fungicides progress has been made along three main lines; by using chemicals which are translocated within the plant and are themselves fungitoxic; by using chemicals which are broken down by the plant or the pathogen to form systemically distributed fungicides; and by using compounds which are neither fungicidal *per se* nor give rise to fungicidal degradation products but interfere with the host's metabolism and render it resistant to attack. Prof. van der Kerk reported on the progress being made in Holland along all three of these lines. Procaine (diethylaminoethylester of *para*-aminobenzoic acid) shows promise as a systemic fungicide *per se*, but none of its derivatives is of interest and it may be significant that procaine has so far been shown to be active only against powdery mildew on cucumber. This disease again showed itself to be useful for the demonstration of systemic fungicidal activity with sodium dimethyldithiocarbamate. This compound, which is an active plant-growth regulator, is degraded within the bean plant and, by means of an elegant bioassay technique involving *Glomerella cingulata*, can be shown chromatographically to form at least three antifungal derivatives. Resistance to *Cladosporium cucumerinum* has been increased by providing cucumber plants with enantiomorphs of naturally occurring amino-acids; likewise, thiourea and phenylthiourea, which are inactive *in vitro*, have been found to confer resistance against certain diseases when provided to plants through their roots.

The activity of fungicides in relation to their chemical structure was considered by several speakers in relation to the particular compounds with which they were interested. Dr. D. Woodcock, in his contribution, dealt specifically with structure-activity correlationships of some nitrogenous antifungal chemicals. The importance of physical properties such as lipophilic character was discussed in relation to mode of action. Chelation was also considered, in particular with regard to its influence on the fungitoxicity of oxine (8-hydroxyquinoline) and related compounds. Other compounds dealt with by Dr. Woodcock included imidazolines, guanidines and triazines.

Prof. R. L. Wain and his colleagues at Wye College have been closely associated with research on systemic fungicides for some years and Prof. Wain gave an account of the progress made in this field. He directed attention to the important fact that most plants possess a wide degree of resistance to fungal attack, and pointed out that in some cases this resistance may depend on the possession by the plant of anti-fungal chemicals within its tissues. The observation that the tissues of *Vicia faba* were fungitoxic provided an opportunity of investigating this system from the point of view of "natural systemic fungicides". The isolation of the anti-fungal compounds present in *Vicia faba* has, in fact, been in progress at Wye for some time. Early in these investigations it had to be shown that the observed effect was due to the presence of fungitoxic chemicals and that the broad bean was the most suitable subject for the investigation. Details of the biological work leading to chemical extraction of the fungal inhibitors and the techniques developed for dealing with large numbers of fractions obtained from the extracts were given by Dr. D. M. Spencer. Dr. C. H. Fawcett then described the successful extraction of the most potent two chemicals ; one a keto-yne and the other a sulphur-containing acid with certain properties similar to those of a nucleotide. While somewhat unstable, there is no doubt that these materials are highly fungitoxic and their presence in *Vicia faba* may well prove to be of considerable significance.

The last two contributions dealt with two new fungicides now in use. D. J. Higgons described the research which led to the field trials and ultimate commercial application of 2,6-dichloro-4-nitroaniline, particularly against *Botrytis cinerea* on lettuce, and Dr. W. F. Jepson dealt with the orchard use against scab diseases and the possible mode of action of *n*-dodecylguanidine acetate.

The contributions to this symposium ranged widely within the field of fungicides. The practical aspects of their use were discussed, theories on mode of action were described, research being carried out towards the development of new fungicides was reported and details were given of two new materials now on the market. Perhaps part of its undoubted success can be ascribed to the comparative rarity of meetings such as this and it is to be hoped that others will follow at suitable intervals. The organizers of the symposium are to be congratulated on their choice of subject, and the participants—indeed all those interested in fungicides—look forward to the early publication of the monograph containing the contributions and discussions. D. M. Spencer

THE NATIONAL PARKS COMMISSION

THE eleventh annual report of the National Parks Commission*, which covers the year ended September 30, 1960, again records disappointment at the non-introduction in Parliament of any Bill to give effect to the amendments to the Act which the Park Planning Authorities and the Commission recommended to the Minister. This was in spite of the fact that at a Conference of Park Planning Authorities at Windermere in May, the Permanent Secretary to the Minister stressed the importance of having suggestions ready in case they were wanted at short notice. Within the limits of its powers, the Commission records a year of quiet progress, particularly in the designation of areas of outstanding natural beauty, for which the Minister has now confirmed 12 orders covering 1,700 square miles, and several local planning authorities have taken advantage of the special powers provided by the Act for actions aimed at preserving and enhancing the natural beauty of these areas. After informal discussions with the chief bodies concerned the Commission is now considering what advice to give to the Government regarding the Broads in the light of the recommendations of the Bowes Committee of inquiry into inland waterways.

Much of the report consists of notes on progress and activities in the individual Parks, which it is impossible to summarize here, but with the excellent illustrations which continue to characterize the report, the Commission's annual report is itself a first-class piece of publicity, and one encouraging general note struck in the report is that it has at last been able to expand such activities and to appoint a small additional staff for the purpose. A conference of park authorities on the subject was arranged in July 1960, at which one topic discussed was the provision of information centres in all the National Parks. This has been limited hitherto primarily by finance which the 1949 Act failed to provide. The Commission regards such information centres as of the utmost importance and has proposed to the Ministry of Housing and Local Government an increased appropriation for the purpose.

A disappointing feature of the Commission's work stressed in the report is the lack of progress with long-distance routes. Although all but one of seven proposals for such routes submitted to the Minister have been approved, after ten years work not a single route has yet been completed on the ground, and there are still major difficulties in regard to the earliest and longest—the Pennine Way. Apart from this, development questions figure largely in the preoccupations of the Commission, the question of electricity distribution lines underground being again prominent. Strong exception is taken to the attitude of the South Wales Electricity Board to a request by the Pembrokeshire County Council for a small amount of underground cable in the vicinity of Whitesands Bay, St. David's, as quite at variance with the assurances given by Lord Mills when Minister of Power in a debate on the Electricity Bill, 1957. Moreover, in spite of further assurances given on the amenity clause in the Act in a debate in the House of Lords, in February, the Commission, while noting the Government's claim in the debate that all officers of the Electricity Boards are fully alive to the provisions of Section 37 of the Act, suggests that their appreciation of its implications could sometimes be improved.

Like difficulties have arisen in electricity supply to Borrowdale and to Martindale, and while regretting the existence of the Ballistic Missile Early Warning Station at Fylingdales in the North York Moors National Park, the Commission records its thanks to the Air Ministry for collaboration in an attempt to minimize the impact on the scenery. Protests was also made against the proposal, now abandoned, to rebuild Princetown Prison in the Dartmoor National Park and the Peak Park Planning Board was supported in its opposition to the proposal of the Sheffield Corporation to construct a refuse tip at Ringinglow, a proposal finally rejected by the Minister on appeal. Much anxiety is also expressed about the multiplicity of proposals for the erection of aerial masts on prominent sites, and despite the existence of Government arrangements, under which all proposals for new masts must be cleared centrally, and anyone proposing a separate site must show strong reasons for not conforming, the Commission continues to watch the position with some concern, although permission to erect a police mast of 150 ft. on Foel Eryr in Mynydd Preseli has recently been refused by the Minister.

* National Parks and Access to the Countryside Act, 1949. Eleventh Report of the National Parks Commission for the year ended September 30, 1960. Pp. vi+71+8 plates. (London: H.M.S.O., 1961.) 6s. net.

THE WORLD HEALTH ORGANIZATION

THE outstanding activity of the World Health Organization in 1960 was the assistance given to enable the Republic of the Congo (Leopoldville) to meet the emergency health situation in which it found itself immediately after attaining independence. This is described in the annual report of the Director-General for 1960*.

Acting on a request from the Secretary-General of the United Nations for assistance with the rapidly deteriorating health conditions in the Congo, the Organization accepted the responsibility of co-ordinating, through the Ministry of Health, the activities of all international personnel concerned with the health of the civilian population. The report outlines the action taken by the World Health Organization during July, August and September, with the assistance of medical teams provided by national Red Cross societies and certain Governments.

The programme for the eradication of malaria from the world is reported as having made significant progress in various fields, although handicapped by a persistent shortage of funds. Sixty-one countries

* Official Records of the World Health Organization, No. 105: the Work of WHO, 1960—Annual Report of the Director-General to the World Health Assembly and to the United Nations. Pp. x+224. (Geneva: World Health Organization; London: H.M.S.O., 1961.) 6 Swiss francs; 10s.; 2 dollars.

are now fully engaged in this programme; in another nineteen, planning has reached an advanced stage, and, in seventeen other areas—mainly in Africa—the Organization is assisting with preliminary work. Some idea of the complexity and scope of these campaigns is conveyed by the variety of the Organization's work on malaria as reflected in this report—a wide range of research, assistance with training, planning and organizing campaigns and, as the programmes advance, with evaluating and assessing their efficacy and results.

The World Health Organization's assistance to Governments for the control of yaws, smallpox and tuberculosis, leprosy and various diseases increased significantly during the year. Medical research was intensified on practically all communicable diseases, the main co-ordinated effort being centred on virus diseases and on bilharziasis.

The year 1960 was the first year of full implementation of the intensified programme of medical research. Although the programme is concerned mostly with communicable diseases, work has begun on cancer, cardiovascular diseases, radiation medicine and human genetics. It is made clear in the report that the Organization's role is primarily to provide services to research and to co-ordinate the work being done in national centres. "It is, I believe", writes the Director-General in his introduction, "a gratifying fact that much of the medical research supported by WHO has grown out of the Organization's current field programme and is being carried out by national institutes and national staffs".

For the World Health Organization, as for the United Nations, the emergence of new independent States brought a marked increase in membership. In planning for the future, account has been taken of the particular needs of these newer countries, so as to ensure the most effective use of their often limited resources in the progressive development of balanced health programmes.

In most countries essential health work is being handicapped by shortage of trained professional and auxiliary personnel, and much of the Organization's work is concerned with education and training in one form or another. Under the Organization's fellowships programme, more than 1,000 fellowships were provided in the period covered by the report. In addition to details of the fellowships awarded for both group training and individual studies, the report gives information on the other aspects of the World Health Organization's work to further the training of professional and auxiliary health workers: courses, study tours, seminars and a variety of other meetings were organized, and experts were provided to serve as visiting teachers or as members of demonstrations and training teams. For example, in the first nine months of 1960, the Organization sent more than 160 nurses to help with nursing and midwifery training in forty-five countries.

In the field of environmental sanitation, emphasis was laid on the training of sanitary engineers and sanitarians, and on providing safe water-supply and waste disposal. In certain regions—Europe, for example—work was concentrated on river, lake and sea pollution and other problems resulting from the rapid growth of cities, such as air pollution. The growing use of atomic energy in power production, and of ionizing radiation in industry and medicine has given rise to several problems, and many Governments are becoming increasingly aware of their responsibilities in radiation health. The Organization's programme is orientated mainly towards assisting health authorities to strengthen their competence in radiation questions in order to meet these responsibilities.

MEASURING INDIGESTION

INVESTIGATORS in the food research group and analytical chemistry laboratories of the Stanford Research Institute have developed a method to measure the degree of digestibility of any food for a particular individual. Basically, one reason for the discomfort caused by certain foods is related to the bacteria flora that inhabit the intestines of all mammals. When a person eats a food he can digest only partially it is attacked, when it reaches the lower part of the small intestine, by organisms which digest the remaining material. These bacteria form hydrogen gas as a by-product in the process, and resulting discomfort depends on the amount of gas; this, in turn, depends on the constitution of the flora of that individual and the quantity of undigested food.

The project team reasoned that if gas pressure is present, the hydrogen should pass through the semi-permeable intestine wall into the blood stream. Because hydrogen is inert and therefore unchanged by body chemistry, the gas should eventually pass into the lungs and be expelled in the breath. They modified a gas chromatograph so that, if a bag is blown up by the subject, the hydrogen in the breath can be measured. By making the test after feedings of separate foods, they could determine what foods and how much are causing indigestion.

Some aspects of the investigation were sponsored by the U.S. Department of Agriculture and the Idaho Bean Commission. A unique application of the method was utilized by the latter organization and is outlined in a recent issue of *Research* (13, No. 1; January–February 1961). Beans in many individuals produce hydrogen and methane, which also are detectable by chromatography. A chemical and leaching process was developed by the food research group to make beans more generally digestible and greatly reduce gas formation, while not altering their taste. The breath-analysis technique was a useful tool in proving the bean treatment.

The individual digestive idiosyncrasies of animals, which cannot announce the fact that they have 'heart-burn', may be detected by the chromatograph method. Various medical applications have also been suggested. For example, because intestinal bacteria populations increase rapidly by feeding on foods undigested in the stomach, the chances for future digestion problems are increased. With individual analysis, it may become possible to control the number of undesirable bacteria through selection of easily digested foods which will 'starve them out'.

A RADIO WAVE MECHANISM TO ACCOUNT FOR THE KNOWN DISTRIBUTION OF VAN ALLEN BELTS ABOUT THE EARTH

By JOSEPH M. BOYER

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A NUMBER of recent theories[1,2] related to the entrapment of charged particles in the Van Allen belts about the Earth appeal solely to the magnetostatic field of the Earth or Sun for the 'bottle' or trap mechanism. It may be of some value, conversely, to ascertain if the Van Allen belt phenomenon can be predicted in some detail by a simple theory which deliberately omits any recourse to the magnetostatic flux of the Sun or Earth as a charge-trapping means.

Due to the work of Mie[3] and Debye[4] it may be shown that upon the closed surface of a spherical body of matter a system of resonant modes of current will be established by illumination of the body by incident electromagnetic waves vibrating at frequencies within a certain portion of the spectrum called the 'critical band'. Excitation of such resonant surface current modes on the spherical body will, in turn, produce maxima in the scattered electromagnetic fields in the spherical directions φ, θ, of the space surrounding the body; such field distributions will differ from those established by the same body when illuminated by waves having vibrational frequencies lying outside the 'critical band' spectrum region. Here, the electrical constant of the matter comprising the spherical body is denoted k_1, and the electrical constant k_2 of the immersion medium about the body is first assumed to be that of a vacuum. We have: $k_1\epsilon = \epsilon_1/\epsilon_0$; $k_1\mu = \mu_1/\mu_0$; $k_2\epsilon = \epsilon_0 = 8{\cdot}854 \times 10^{-12}$ farads/metre (M.K.S.); $k_2\mu = \mu_0 = 1{\cdot}257 \times 10^{-6}$ henries/metre (M.K.S.); and k_1/k_2 is other than unity in terms of the critical band frequency spectrum considered.

Now, let the medium k_2 contain a continuous spectrum of electromagnetic energy emitted by a source so distant that wave-fronts are essentially plane in the vicinity of the sphere. By the spherical vector wave-functions, the resultant electromagnetic field will be:

$$E = -\sum_n (a_n M_n + b_n N_n), \quad \bar{H} = -\sum_n \frac{k}{i\omega\mu}(a_n \bar{N}_n \bar{M}_n) \quad (1)$$

The particular solution of equation (1) for the sphere is well known; therefore the reader is referred to the excellent treatment of the series by J. A. Stratton[5] for details. By means of equation (1) we can secure the relationship of wave-length λ to sphere radius a for the first three significant resonance modes. These are:

No. 1. $\alpha = 2\pi/\lambda_a = 1{\cdot}0$
$\lambda = 6{\cdot}28a$

No. 2. $\alpha = 2\pi/\lambda_a = 2{\cdot}30$
$\lambda = 2{\cdot}73a$

No. 3. $\alpha = 2\pi/\lambda_a = 3{\cdot}45$
$\lambda = 1{\cdot}82a$

using the wave-number $2\pi/\lambda$ in standard notation.

Permit the Earth, stripped of its atmosphere, to be considered the spherical matter zone immersed in space. A line x is drawn from the centre of the Earth along the ecliptic plane to terminate at a point o. The length of the line x is $149{\cdot}5 \times 10^6$ km. The source of incident waves is located at point o. For the Earth sphere with a mean radius equal to 6,371·23 km., the first three resonant modes may now be ascertained:

No. 1. $\lambda = 40{\cdot}01 \times 10^6$ m. = 6·28 Earth radii. Frequency = 7·49 cycles/sec.

No. 2. $\lambda = 17{\cdot}39 \times 10^6$ m. = 2·73 Earth radii. Frequency = 17·25 cycles/sec.

No. 3. $\lambda = 11{\cdot}59 \times 10^6$ m. = 1·82 Earth radii. Frequency = 25·88 cycles/sec.

Effects which take place along the line x as the result of electromagnetic resonances of the illuminated Earth are of first interest. For the case of Mie scattering of the Earth at modes 1, 2 and 3, we can determine the magnitude of the secondary field established in the direction of o, compared with a fictitious isotropic scattering body which maintains a constant field E^s in all spherical directions φ, θ, when radiating a total power P^s isotropic. The scatter field magnitude in the direction o is called backscatter[6] cross-section. Fig. 1 shows the backscatter cross-section σ of a highly conducting sphere (solid line) and that of a water sphere (dotted line) normalized with respect to πa^2.

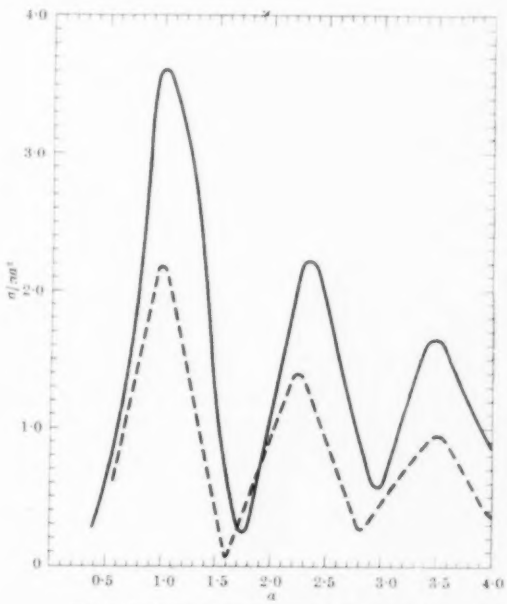

Fig. 1. Backscattering from spheres (from Aden)
a, radius of sphere; $\alpha = Ka = \frac{2\pi}{\lambda} a$
———, Metal sphere; — — —, water sphere

It will be noted that the backscatter cross-section of a sphere illuminated with energy vibrating at frequencies within a critical bandwidth, approximately $\alpha = 0{\cdot}8$ to $\alpha = 6{\cdot}0$, differs markedly from the case of either Rayleigh or specular scatter. Within the critical band, it is seen that peaks and minima are evident in the magnitude of the backscatter cross-section. Again, Fig. 1 discloses that although the electrical constant of the matter making up the spherical body may affect the magnitude of backscatter cross-section it does not sensibly disturb the radius/wave-length relationship of the electromagnetic resonances. The energy scattered back in the direction o will establish electromagnetic standing waves[7] along this axis due to phase interference with the incident field. The intensity of the maxima (loops) of such standing waves will be greater for those wave-lengths within the critical band producing resonance conditions with the Earth sphere, although all finite magnitudes of backscatter field from the Earth will introduce some standing wave phenomenon for frequencies within the illumination spectrum. Emphasis is placed, however, on the very lowest frequency resonances as a decelerating mechanism for high-energy particles.

A. L. Aden[8] has demonstrated the existence of electromagnetic backscatter standing waves produced by water and metal spheres in experiments conducted at a wave-length of 16·23 cm. As the scattered field from the Earth is a spherical wave in the vicinity of the planet, the standing wave which results is damped by radiation. Its configuration, for a single frequency, is therefore a damped sinusoidal wave[9] along the line x, the position of the loops and nodes stationary, with an amplitude variation at a period ω. It must be admitted that employment of an Earth stripped of its atmosphere envelope introduces some grounds for objection to the realism of the model offered so far. In this cursory survey of the theory, only an approximation of the true radius of the Earth scattering shell will be made, for it is clearly evident that rigorous treatment will display a frequency selective term for the extremely low-frequency scatter radius, with complicated entrapped wave effects occurring at the high-frequency end of the spectrum.

The Lyman–Spitzer analysis[10] of the planetary atmosphere shell is selected as a model. The Spitzer atmosphere acts like a gas up to about 600 km., with lighter gases predominating in the atmosphere population above about 300 km. Although free electron densities quoted by authorities differ to some degree, sufficient accuracy here will be attained by use of a density curve with peaks at about 300 km.[11], then decreases at higher altitudes. A mean scattering shell altitude of 300 km. will be selected, producing a total sphere radius of 6,671·23 km. The atmosphere 'corrected' Earth will then introduce resonance modes in the illuminating spectrum at the following wave-lengths:

No. 1. $\lambda = 41{\cdot}89 \times 10^6$ m. = 6·57 Earth radii. Frequency = 7·16 cycles/sec.

No. 2. $\lambda = 18{\cdot}21 \times 10^6$ m. = 2·86 Earth radii. Frequency = 16·47 cycles/sec.

No. 3. $\lambda = 12{\cdot}14 \times 10^6$ m. = 1·9 Earth radii. Frequency = 24·71 cycles/sec.

I am confining attention in this article to the electromagnetic standing waves along line x generated by scatter interference only at wave-lengths Nos. 1, 2 and 3. The position radially from the Earth for each standing wave distribution may be computed or taken directly from the Aden measurements given in the reference. There will be no interaction between such individual frequency fields existing along x if we assume initially that the space k_2 surrounding the planet is free of all charges. This pure field or fictitious state will be labelled the particle-free standing wave case. Under this condition, examination of the polarization characteristics of the incident and scattered wave shows that the conditions for maximum interference between the two fields can exist only within a narrow, solid angle cone pointing toward the point o, with the magnitude of the induced standing wave distribution falling off in other radial directions φ, θ.

Now, let the radiant source at o not only generate electromagnetic waves but also high-energy charged particles, some of which approach the Earth along the incident axis x. J. R. Coleman[12] has examined my matter-induced electromagnetic standing-wave theory in terms of the electron capture mechanism involved in the radio-frequency case. L. M. Kovrizhnykh and V. H. Veksler[13] have studied a similar situation involving capture oscillations of both charged and neutral particles in a radio standing-wave type cyclotron. These analyses emphasize that for a fixed loop field strength of an electromagnetic standing wave the force F exerted on the particle is proportional to wave-length. In terms of decelerator action, Coleman notes that even in the case of a 'solar wind' electron moving at a velocity of 10^6 m./sec. capture will occur when such particle is injected into a radio standing wave the fundamental wave-length of which lies between 10×10^6 and 60×10^6 m. with a loop field-strength of 0·17 V./m. Such extreme particle velocity, of course, is not required in the study of the Van Allen belts found near the Earth, but assumes importance in terms of wave-length/field strength requirements for much larger planets such as Jupiter or Saturn, the first-mode resonance wave-length of which is at least 439×10^6 m. Coleman also makes a preliminary extension of the equivalent black-body temperature of the Sun into the Earth critical spectrum band with encouraging results.

Briefly, then, we have a planetary mass, under illumination from a radiant source of large size such as a star, inducing electromagnetic standing waves along the incident direction. Here, individual frequency components of such standing waves do not interact field-wise but do act on the injected charges in such a manner that an integrated force obtains, with the longest wave components of the stationary wave producing first-order effects. Particles injected into such standing-wave envelope region will either be captured if the mass/velocity relationship to wave-length/field strength is critical or be deflected as if by an electromagnetic lens and escape. The requirement for a mechanism to establish the radial Van Allen belts observed about the Earth is still not satisfied, however, by this model, for appreciable magnitude 'capture' fields in the form of large intensity standing waves do not exist at other than the incident axis direction.

However, now let an injected particle, say, an electron, be captured by the incident axis electromagnetic standing wave. Once captured, the electron enters into a steady-state condition which I shall term the particle-wave case. Such particle, as

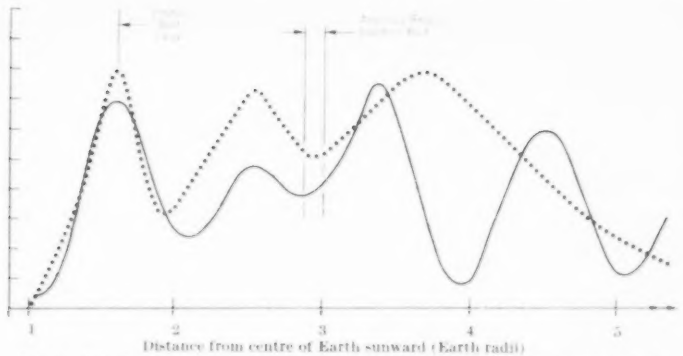

Fig. 2. Distribution of known Van Allen belts compared with calculated wave due to Earth dipole resonance in solar elf electromagnetic illumination
. . . . , Van Allen belt distribution (after Simpson *et al.*); ——, calculated Earth electromagnetic standing wave envelope for modes 1, 2 and 3

Coleman shows, will oscillate along a path concentric with the incident axis x. Studying the Coleman analysis, I realized that by appealing to electromagnetic theory, such an oscillating charged particle becomes a source of current in space. Neglecting the ramifications of the relativistic mass of such oscillating particle in this article, it is clear that the particle becomes a new source of low-frequency radiation in the Earth capture model. Due to the extremely low frequency of the entrapping wave, the particle space current constitutes an extended line source; the Poynting vector of the particle-generated wave must, therefore, lie normal to the incident axis x. Radiation of electromagnetic energy in the direction normal to axis x will introduce increased magnitudes of interference with the sphere scattered wave in a new direction φ, θ displaced off the incident axis. The effect of this interference is to increase the width of the conical standing-wave region pointing to the source. The process is now cumulative in that newly injected particles can be entrapped or else migrate into the widened solid angle zone of standing wave to oscillate along new radial directions. Ultimately, the planet will be surrounded by a set of anular zones of standing waves with loops and nodes distributed radially.

Fig. 2 shows the mathematical average envelope obtained by superimposition of the standing waves resulting solely from resonance modes 1, 2 and 3 of the Earth exosphere scatter model traced as a solid line. The radial distribution of the closest Van Allen belts after Simpson *et al.*[14] is given as a dotted line. It will be seen that a maximum in envelope intensity (loop) exists close to the 1·5 Earth radii position of the known proton belt, and the anomaly in the electron belt at 2·85–3·0 Earth radii is evident without recourse to the Cape Town variation in the Earth magnetostatic field. Beyond 4 Earth radii, the computed field envelope falls off more rapidly than the distribution given by Simpson, with a new maximum produced at about 4·4 Earth radii. It is felt that a model based on the addition of a larger number of critical band frequency components, as well as an extension of the radial distance scale, will fit the known belt distribution more satisfactorily.

Calculations by me of the standing wave distribution in a plane normal to the ecliptic (θ) shows that when the first three resonance modes are superimposed, a deep null or electromagnetic 'pole' occurs at 93 degrees above the radio 'equator' or ecliptic plane of the Earth, permitting deep penetration of particles in the high latitudes.

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OBSERVATION OF FAR-INFRA-RED RADIATION FROM SHOCK WAVES

By M. F. KIMMITT, A. C. PRIOR and P. G. SMITH

Royal Radar Establishment, Malvern

INFRA-RED radiation in the wave-length range 0·1–0·6 mm. has been observed in the emission from a plasma generated by a shock wave in argon. This work follows a successful joint experiment with the Atomic Energy Research Establishment, Harwell, in which the emission in the range of wave-lengths 0·1–1·6 mm. was observed from plasmas produced in *Zeta*. The apparatus used was designed for the latter experiment, the details of which are being published elsewhere[1]. The spectra obtained from the observations on the shock tube described here, as from those on *Zeta*, are characteristic of bremsstrahlung, but show regions where the emission is reduced by the black-body limitation.

Under the conditions of these experiments, and in the wave-length region examined, line and band spectra arising from transitions between atomic energy-levels, or between energy-levels of molecules present as impurities, should be absent or insignificant. The observed emission should therefore be due entirely to free-free electron transitions (bremsstrahlung) which, according to the calculations of Gaunt[2]

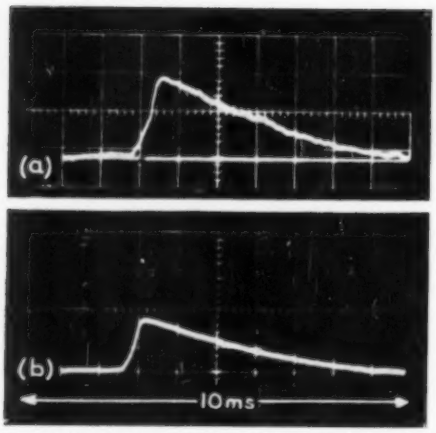

Fig. 1. Oscilloscope traces obtained without spectral resolution. (*a*) Heavily absorbing plasma produced from shock into 1 torr argon. (*b*) Relatively transparent plasma produced from shock into 0·25 torr argon

and Cilhé[3], result in a surface brightness $P_{\text{eff.}}$ given, when $h\nu \ll kT$, by :

$$P_{\text{eff.}}\, d\nu = 1{\cdot}63 \times 10^{-35}\, Z^2\, g\, T^{-1/2}\, n_e n_i\, d\, d\nu \text{ watts/cm.}^2\text{/steradian/cm.}^{-1} \quad (1)$$

where Z is the ionic charge, T the absolute temperature, n_e and n_i the electron and ion densities, d the plasma thickness, ν the wave number, and g the Gaunt correction factor given by :

$$g = (\sqrt{3}/\pi) \ln (3kT/2h\nu c)$$

If we take $n_e = n_i = n$, $Z = 1$, and a mean value of 3 for g, these being appropriate to the conditions of the experiment, (1) reduces to :

$$P_{\text{eff.}}\, d\nu = 5 \times 10^{-38}\, n^2\, T^{-1/2}\, d\, d\nu \text{ watts/cm.}^2\text{/steradian/cm.}^{-1} \quad (2)$$

This expression neglects self-absorption, which will prevent the emission from exceeding the black-body value for the appropriate temperature, T; it also neglects plasma resonance effects, and applies only at frequencies sufficiently above that for plasma resonance.

The radiation emerged through a crystal quartz window of diameter 1·5 cm. in the side of the shock tube of 5 cm. diameter, and passed into the spectrometer, which has been briefly described by Roberts[4]; a cone of radiation of half-angle 12° was accepted through the window. The detector was a carbon bolometer similar to that described by Boyle and Rodgers[5], and had a time constant of about 4 msec., the amplified output being displayed on an oscilloscope. The pass band of the spectrometer is normally about 10 per cent of the wave-length to which it is set, but where a greater signal was required in order to examine pulse shapes, the whole spectrum from 0·1 to 1·5 mm. wave-length could be accepted. The energy sensitivity of the system was calibrated by comparison with a black body of known temperature.

The length of heated gas behind the shock front was about 50 cm. as it passed the window, and its velocity about 5×10^5 cm./sec. ; the corresponding transit time of 100 μsec. was much less than the time constant of the detector, which therefore gave an approximately integrated signal. The shock-tube walls are silvered and would be expected to have a reflectivity of 98–99 per cent. For a heavily absorbing plasma, the signal received from the plasma covering the window and filling the spectrometer aperture is preceded and followed by a signal of about half the value, only half the spectrometer aperture being then filled with radiation. This is demonstrated by Fig. 1(*a*), where the steep central portion of the integrated signal is preceded and followed by portions of lower slope. The transit time of the hot gas as deduced from Fig. 1(*a*) agrees with that obtained independently from measurements by resistance thermometer. For a plasma having negligible absorption, and with perfectly reflecting walls, the radiation received should be independent of the position of the shock wave in the tube, since each part of the plasma will fill one or other half of the spectrometer aperture, for all positions of the plasma in the tube. This is demonstrated in Fig. 1(*b*), where no abrupt changes of slope occur but only gradual changes due to imperfect reflectivity.

In principle, the instantaneous radiation intensity can be deduced by measuring the slopes on photographs such as Fig. 1. In practice, the signal was generally too small for this, and instead the peak amplitude was measured and related to the slope by comparison with traces obtained without spectral resolution, where the slopes could be measured ; the results are plotted in Fig. 2. The shock-wave Mach number was measured by means of resistance thermometers spaced either side of the spectrometer aperture at approximately 1 metre apart. At an initial pressure of 1 torr of argon, a Mach number of 15 was measured. From the theoretical calculations

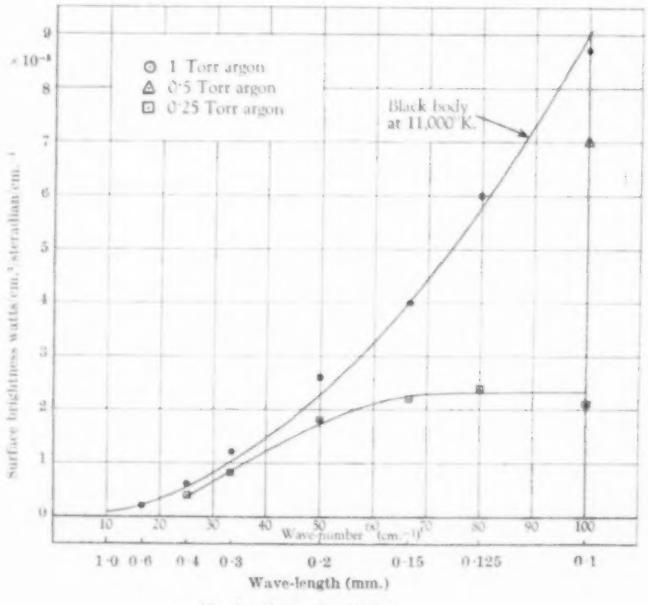

Fig. 2. Observed radiation

of Frood (private communication), these shock-wave conditions should give a temperature of ~11,000°K. and an electron density of $\sim 3 \times 10^{16}$ cm.$^{-3}$, assuming equilibrium to be established in the plasma behind the shock front. For this temperature and density, equation (2) would, for a wave-length of 0·1 mm., give a value of brightness equal to that from a black body if d were put equal to about 1 mm. Thus a black-body spectrum is to be expected, and the experimental points do in fact fit the 11,000°K. black-body curve better than would be anticipated from consideration of likely calibration errors of the spectrometer. Since the available thickness of the plasma, allowing for reflexion from the walls, is ~ 150 cm., any density n greater than ~ 10^{15} cm.$^{-3}$ would be adequate to account for the black-body shape of the spectrum. However, the abrupt changes of gradient in Fig. 1(*a*) already discussed show that the radiation penetrates only a small portion of the plasma, and this raises the lower density limit to about $2\text{–}3 \times 10^{15}$ cm.$^{-3}$.

For an initial pressure of 0·25 torr of argon, the values of temperature and density calculated from the shock-wave parameters are $T \sim 11{,}000$°K. and $n \sim 1 \times 10^{16}$ cm.$^{-3}$. The approximately flat part of the spectrum indicates that the absorption for the short wave-lengths must be small, as confirmed by the pulse shape (Fig. 1(*b*)), so that for this region equation (2) should apply; using this with $d = 100$ cm. (shock length = 35 cm.) a value of $n \sim 6 \times 10^{14}$ is obtained. The observations at the longer wave-lengths give good agreement with the calculated temperature. The single observation with an argon pressure of 0·5 torr being slightly below the black-body curve indicates a value of $n \sim 10^{15}$ cm.$^{-3}$ as compared with 2×10^{16} cm.$^{-3}$ deduced from the shock-wave parameters.

All the measurements, both of shock-wave parameters and of radiation, are rather crude time averages for shocks which were probably decaying at a significant rate. Under these conditions, the agreement of the temperature deductions is remarkably good, and the lower densities deduced from the radiation measurements are reasonable, since the calculated densities are for thermal equilibrium, which may well not be reached.

These exploratory measurements have indicated the potential value of far-infra-red spectroscopy for shock-wave plasma diagnostics. Investigations of plasmas with electron densities greater than ~ 5×10^{14} cm.$^{-3}$, which are beyond the range of standard microwave methods, should be possible, and with a faster detector[6] now being actively developed, much more detailed information should be obtainable than in the present experiments.

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PHOTOPRODUCTION OF HYDROGEN, PHOTOFIXATION OF NITROGEN AND A UNIFIED CONCEPT OF PHOTOSYNTHESIS

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BY fractionating isolated chloroplasts, it has been demonstrated experimentally that the overall process of photosynthesis can be separated into a light and a dark phase[1]. The dark phase includes a series of reactions, driven by adenosine triphosphate and reduced triphosphopyridine nucleotide, that convert carbon dioxide into carbohydrate[1–4]. Since the same series of dark reactions is now known to occur also in non-chlorophyllous cells[5,6], it is appropriate to regard carbon dioxide assimilation as another example of cellular activity which is essentially similar in photosynthetic and non-photosynthetic cells. Consequently, carbon dioxide assimilation must be regarded as lying essentially outside the domain of photosynthesis proper[3]. (This conclusion is based on the fact that grana, as contrasted with whole chloroplasts, cannot assimilate carbon dioxide to the level of carbohydrates but retain a capacity for photochemical oxygen evolution and photosynthetic phosphorylation[4,7]. Our concept would not exclude a *catalytic* participation of carbon dioxide in the mechanism of oxygen evolution as has recently been proposed by Warburg *et al.*[12] and supported by Stern and Vennesland[13].)

What is peculiar to photosynthesis is its light phase. For green plants this comprises the light reactions that produce adenosine triphosphate and reduced pyridine nucleotide[2,7–9]: cyclic photophosphorylation (equation 1) and non-cyclic photophosphorylation (equation 2). The terms 'cyclic' and 'non-cyclic' denote the respective electron-flow mechanisms[10] that we have postulated to explain the experimental findings which are summarized by reactions 1 and 2.

$$n\text{·ADP} + n\text{·}H_3PO_4 \xrightarrow{\text{light}} n\text{·ATP} \qquad (1)$$

$$2\ \text{TPN} + 2\ \text{ADP} + 2\ H_3PO_4 + 2\ H_2O \rightarrow 2\ \text{TPN}H_2 + 2\ \text{ATP} + O_2 \qquad (2)$$

Cyclic photophosphorylation (reaction 1), a reaction that produces only adenosine triphosphate, has been found in every photosynthetic organism tested so far: higher green plants[11–20], algae[21–23] and photosynthetic bacteria[23–25]. This is in agreement with the view presented elsewhere[10,19,20] that cyclic photophosphorylation (reaction 1) is the common denominator of all photosyntheses in plants and in bacteria. By contrast, non-cyclic photophosphorylation, as represented by reaction 2, has thus far been found only in chloroplasts.

The assimilation of carbon dioxide to the level of carbohydrate requires, in addition to adenosine triphosphate, reduced pyridine nucleotide. In iso-

lated chloroplasts, these joint requirements for assimilation of carbon dioxide are satisfied by a combination of cyclic and non-cyclic photophosphorylation[2] (reactions 1 and 2). In photosynthetic bacteria, we have previously investigated only the case in which the electron donor was hydrogen gas. The contribution of light was then limited to the formation of adenosine triphosphate by cyclic photophosphorylation (reaction 1); reduced pyridine nucleotide was formed in a 'dark' reaction, at the expense of hydrogen gas[29]. We had no experimental evidence previously for a non-cyclic photophosphorylation mechanism in photosynthetic bacteria which would be analogous to non-cyclic photophosphorylation *in* chloroplasts.

The purpose of this article is to present a unified concept of photosynthesis that includes both cyclic and non-cyclic electron-flow mechanisms in photosynthetic bacteria and green plants. Evidence for the operation of a special non-cyclic electron-flow mechanism (without oxygen evolution) in photosynthetic bacteria was obtained by measuring not only photoreduction of pyridine nucleotides[31,32], but also photoproduction of hydrogen and photofixation of nitrogen gas.

Non-cyclic Electron-flow in Bacteria

(a) Photoproduction of hydrogen gas from thiosulphate. Photoproduction of hydrogen gas by photosynthetic cells was first observed by Gaffron and Rubin in the green alga *Scenedesmus*[34] and by Gest and Kamen in photosynthetic bacteria[35]. In algae, the photoproduction of hydrogen seemed to depend on internal electron donors of metabolic origin[33], whereas photoproduction of hydrogen by photosynthetic bacteria appeared to depend on the presence of exogenous organic acids and carbon dioxide[34–40]. The hydrogen evolved has been thought to arise from a photodecomposition of water[33,40] or from a decomposition of α-ketoglutaric acid[38].

From the point of view of the electron-flow theory of photosynthesis[10,41], which does not involve a photolysis of water, the photoproduction of hydrogen could be viewed as a reduction of H^+ by a hydrogenase with the aid of electrons expelled from excited chlorophyll molecules[41,42]. In accordance with that theory, electrons expelled from chlorophyll would be replaced by electrons donated by cytochromes, which would then become oxidized. The oxidized cytochromes would in turn become reduced by the external electron donors. The cytochrome system in photosynthetic bacteria would thus be a gateway for the entry of electrons and for their transfer to chlorophyll, where they would be raised, at the expense of the energy of absorbed light, to a potential sufficient to reduce H^+ (cf. refs. 41 and 42).

If this interpretation were correct, then photoproduction of hydrogen by photosynthetic bacteria should occur at the expense of simple, physiologically suitable, inorganic electron donors—a possibility for which there was no experimental evidence[37]. We therefore undertook to investigate the photoproduction of hydrogen from thiosulphate by *Chromatium* cells.

Fig. 1 shows that *Chromatium* cytochromes are reduced by thiosulphate, and can serve, therefore, as a point of entry for electrons from that source. Fig. 2 shows that in the light, but not in the dark, gas was evolved by *Chromatium* cells in the presence

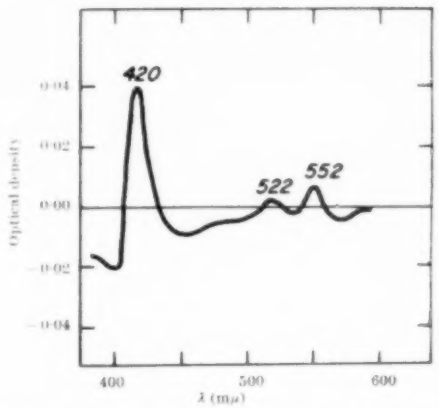


Fig. 1. Reduction of *Chromatium* cytochromes by thiosulphate in a cell-free system. Reaction mixture included, in a final volume of 3·0 ml., 0·1 *M tris* buffer, *p*H 7·8; chromatophores containing 0·1 mgm. bacteriochlorophyll and supernatant fluid corresponding to 0·3 mgm. bacteriochlorophyll. 20 μmoles of thiosulphate were added to one of a pair of Thunberg-type cuvettes and the resulting difference spectrum was measured in a Cary spectrophotometer after 20 min. at room temperature. Gas phase, argon.

of thiosulphate. No carbon dioxide was supplied and potassium hydroxide was present in the centre well of the reaction vessel. Evolution of gas ceased when the light was turned off and was resumed when light was turned on again. The gas evolved was identified as hydrogen by adsorption on palladium asbestos[42]. The photoproduction of hydrogen gas was dependent on the concentration of thiosulphate, and was abolished by heating the cells (Fig. 3). The photoproduction of hydrogen gas approached the stoichiometry of reaction 3, that is, for one molecule of thiosulphate used, two electrons and two protons were combined with the aid of hydrogenase and evolved as hydrogen gas.

The evolution of hydrogen by illuminated *Chromatium* cells showed a marked dependence on *p*H (ref.

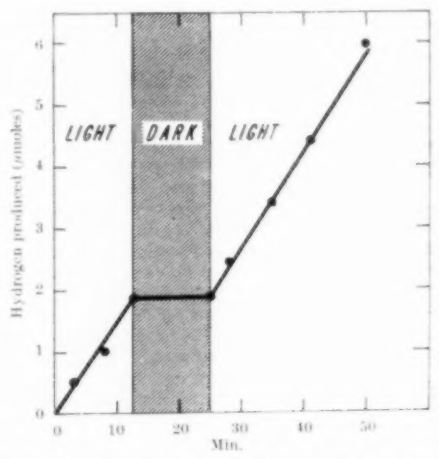


Fig. 2. Light-dependent evolution of hydrogen gas from thiosulphate by *Chromatium* cells. The reaction mixture included 0·1 gm. of washed cells, suspended in 2·6 ml. of a modified nutrient solution from which nitrogen compounds were omitted, 0·3 ml. of 0·5 *M tris* buffer, *p*H 7·2, and 0·1 ml. of 0·2 *M* sodium thiosulphate. 0·1 ml. of 20 per cent potassium hydroxide was put in the centre well of the Warburg manometer flasks. The reaction was run at 30° C. Gas phase, argon. Illumination 50,000 lux

42). The reaction was very vigorous at *p*H 6·5, but little hydrogen was evolved at *p*H 8·0.

Photoproduction of hydrogen from thiosulphate was inhibited by nitrogen gas and ammonium ions[42], as is the evolution of hydrogen in the presence of organic electron donors[21,38]. However, neither ammonia nor nitrogen gas inhibited fixation of carbon dioxide by illuminated *Chromatium* cells when thiosulphate was the electron donor. This indicated that, although under the conditions of our experiments ammonia and nitrogen inhibited the flow of electrons to H^+, they did not inhibit the flow of electrons that is required for assimilation of carbon dioxide by way of pyridine nucleotide.

Growing *Chromatium* cells were found, by analysis, to have oxidized in four days 27 millimoles of added thiosulphate to 54 millimoles of sulphate. During this period the appearance of the culture indicated a transitional formation of elemental sulphur. The results are in agreement with the following sequence of reactions, in which thiosulphate is the donor of electrons that are activated by light and used either for the assimilation of carbon and nitrogen into cellular substance or for the photoproduction of hydrogen gas:

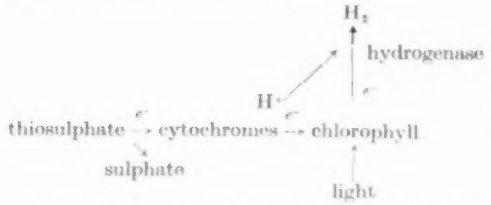
$$S_2O_3^{=} + 2\ OH^- \xrightarrow{\text{light}} S + SO_4^{=} + 2\ e^- + H_2O \quad (3)$$

$$S + 8\ OH^- \xrightarrow{\text{light}} SO_4^{=} + 6\ e^- + 4\ H_2O \quad (4)$$

$$10\ H_2O \xrightarrow{\text{dark}} 10\ H^+ + 10\ OH^- \quad (5)$$

$$\text{Sum: } S_2O_3^{=} + 5\ H_2O \xrightarrow{\text{light}} 2\ SO_4^{=} + 8\ e^- + 10\ H^+ \quad (6)$$

The proposed electron-flow mechanism[42] for the photoproduction of hydrogen from thiosulphate envisages the following electron transfer:



The results with thiosulphate provided the first experimental evidence for hydrogen evolution dependent on light, from an 'inorganic' electron donor by a photosynthetic organism. Similar results were obtained when thiosulphate was replaced by succinate as electron donor[41].

It appears, therefore, that *Chromatium* uses light energy to raise electrons, supplied by thiosulphate to chlorophyll, via cytochromes, to a reducing potential at least equal to that of molecular hydrogen. We have also observed evolution of hydrogen gas in the *dark* by cell-free *Chromatium* preparations when the electron donor was dithionite, a substance that provides electrons capable of reducing protons without requiring additional energy (a catalytic amount of benzyl viologen was used as an electron carrier in this dark reaction).

(*b*) *Photofixation of nitrogen gas.* Nitrogen fixation by photosynthetic organisms (refs. 45, 35, 38, 46–49) may also be viewed as resulting from a non-cyclic electron-flow in which electrons pass from an external electron donor, via cytochromes, to chlorophyll excited by light, and thence to molecular nitrogen.

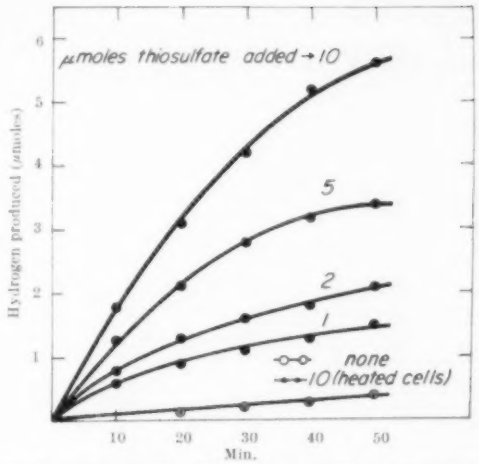


Fig. 3. Photoproduction of hydrogen gas by *Chromatium* cells as a function of added thiosulphate. The control vessel contained cells that had been heated at 100° C. for 10 min. Other experimental conditions are as described for Fig. 2

This interpretation was substantiated by using thiosulphate and succinate as electron donors for fixation of nitrogen gas by illuminated *Chromatium* cells[50]. Fig. 4 shows the result of photofixation of nitrogen gas with thiosulphate as the electron donor. Similar photofixation of nitrogen was obtained with succinate as the electron donor. In both cases, *Chromatium* cells fixed nitrogen only in light. Fixation ceased when the light was turned off and was resumed when the light was turned on again. The dependence of nitrogen fixation on light and an external electron donor was confirmed by the use of nitrogen-15 isotope (Table 1). We are indebted to Dr. C. C. Delwiche for the determinations of nitrogen-15.

In the case of thiosulphate, photofixation of nitrogen was greatly increased by the addition of oxalacetate (Fig. 4), which probably acted as an amino-group acceptor. Succinate served both as electron

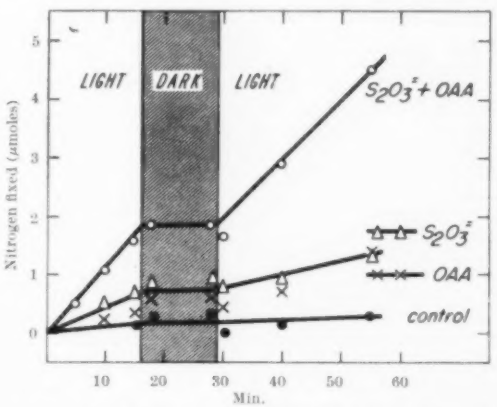


Fig. 4. Effect of thiosulphate and oxalacetate on photofixation of nitrogen gas by *Chromatium* cells. The reaction mixture included in a final volume of 2·9 ml., 0·1 gm. of washed cells, suspended in a modified nutrient solution, *p*H 7·2 (from which nitrogen compounds were omitted). 20 micromoles each of thiosulphate and oxalacetate were added as indicated. 0·1 ml. of 20 per cent potassium hydroxide was put in the centre well of the Warburg manometer flasks. The reaction was run at 30° C. Gas phase, nitrogen. Illumination 50,000 lux

Table 1. Effect of Light and Electron Donors on Fixation of Nitrogen Gas by *Chromatium* Cells

Treatment	Nitrogen-15 atom per cent excess
Dark, succinate	0·004
Dark, thiosulphate + oxalacetate	0·003
Light	0·288
Light, succinate	1·049
Light, thiosulphate + oxalacetate	1·467

40 micromoles each of thiosulphate, oxalacetate and succinate were added as indicated. Other experimental conditions as in Fig. 4, except that no potassium hydroxide was included, and the gas atmosphere was 90 per cent argon and 10 per cent nitrogen containing 30 atom per cent excess nitrogen-15. Reaction time was 2 hr.

donor and as a source of the carbon skeleton that is needed for accepting an amino-group.

In preliminary experiments, when cell-free *Chromatium* preparations were supplied in the 'dark' with a mixture of hydrogen and nitrogen gas, they absorbed more gas than in the control treatment in which the gas phase consisted solely of hydrogen. This seems to indicate that cell-free *Chromatium* preparations can fix nitrogen, with the aid of hydrogen gas as the electron donor—an interpretation that was in agreement with experiments using nitrogen-15 (Table 2). Photofixation of nitrogen-15 by cell-free *Chromatium* preparations, although low by comparison with that by whole cells (Table 1) was above the level of significance of isotopic identification with the mass spectrometer used.

Table 2. Nitrogen-15 Fixation in the Dark by Cell-free Extracts of *Chromatium*

No.	Reductant added	Nitrogen-15 atom per cent excess Experiment *A*	Experiment *B*
1	None	0·0255	0·0274
2	Reduced diphosphopyridine nucleotide	0·0480	
3	Hydrogen gas	0·0362	0·0430

The reaction mixture contained in a final volume of 3 ml.: cell-free extract, containing 0·3 mgm. bacteriochlorophyll, and the following in micromoles: *tris* buffer, pH 7·8, 100; magnesium chloride, 5; benzyl viologen, 0·2. Vessels 1 and 3 received 0·5 μmole diphosphopyridine, and vessel 2, 1 μmole reduced diphosphopyridine nucleotide. All vessels received 0·5 atmosphere nitrogen containing 90 atom per cent excess nitrogen-15. Treatment 3 received in addition 0·5 atmosphere hydrogen gas. The experiment was run at 25° C. for 2 hr. in the dark.

These findings appear to support the view that the role of light in photofixation of nitrogen by *Chromatium* is to generate electrons with a reducing potential that is at least equal to that of hydrogen. When exogenous hydrogen was supplied in the gas phase in the dark, light was no longer necessary for the fixation of nitrogen. Pyridine nucleotides seemed to mediate the reduction of nitrogen. However, the experiments on nitrogen fixation by cell-free *Chromatium* preparations are at an early stage and it would be premature to draw final conclusions.

(c) *Photoreduction of pyridine nucleotides.* The photoreduction of diphosphopyridine nucleotide was observed by Frenkel[31] and Vernon and Ash[32] in *Rhodospirillum rubrum*, and by Ogata *et al.*[51] in *Chromatium*. In more recent experiments we have found that in the presence of succinate and light, unwashed chromatophores from *R. rubrum*, unaided by enzymes from chloroplasts (cf. ref. 32), reduce both di- and tri-phosphopyridine nucleotide.

We have previously emphasized[10,29] that *Chromatium* is capable of reducing pyridine nucleotide in the dark, when supplied with exogenous electron donors, such as hydrogen gas, which yield electrons at the redox-level of pyridine nucleotide. The reduction of pyridine nucleotide is dependent on light when photosynthetic bacteria are supplied with exogenous electron donors, such as succinate, which yield electrons at a redox-level insufficient for reducing pyridine nucleotide without an additional input of energy. It should be noted, however, that in the case of succinate, the relatively low redox potential characterizes only the first oxidation step, that to fumarate. As fumarate is transformed to malate, *Chromatium* can use it to reduce pyridine nucleotide in the dark[29].

Experiments with cell-free systems of *R. rubrum*, to be reported elsewhere (Nozaki, M., Tagawa, K., and Arnon, D. I.), indicate that the non-cyclic electron transport in photosynthetic bacteria that results in pyridine nucleotide reduction is coupled with formation of adenosine triphosphate, thus providing a parallel to non-cyclic photophosphorylation in chloroplasts[7–9]. Experiments are now under way to determine whether the formation of adenosine triphosphate is also coupled with the non-cyclic electron transport that results in the photoproduction of hydrogen or photofixation of nitrogen gas.

To recapitulate, the reduction of pyridine nucleotides, the fixation of nitrogen and the evolution of hydrogen from such electron donors as thiosulphate or succinate are reactions that in photosynthetic bacteria have been found to require an input of light-energy. These photochemical reactions suggest the existence of a non-cyclic electron-flow mechanism in photosynthetic bacteria as represented diagrammatically in Fig. 5.

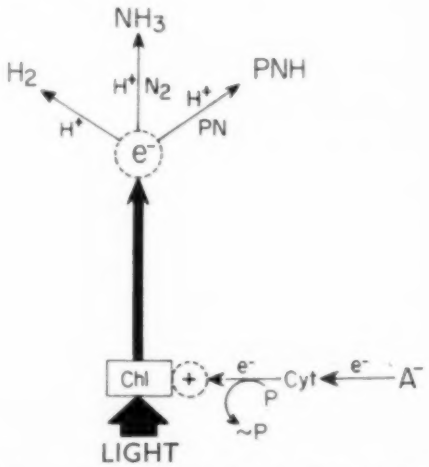


Fig. 5. Scheme for non-cyclic electron flow in *Chromatium*. Details in the text

Non-cyclic Electron Flow and the Evolution of Photosynthesis

Until recently, cyclic photophosphorylation was the only experimentally demonstrable common denominator of the mechanism of photosynthesis in plants and bacteria. Now it has become clear that photosynthetic bacteria may also use part of the absorbed light-energy for a non-cyclic photophosphorylation that is analogous in some respects to that found in chloroplasts.

The most characteristic difference between the non-cyclic electron-flow mechanism of chloroplasts (reaction 2) and bacteria (Fig. 5) is in the electron donor system. In chloroplasts the electron donor is water (that is, OH^-), whereas bacteria cannot use

water but employ inorganic or organic electron donors such as thiosulphate or succinate[52,53].

This interpretation of the difference between the non-cyclic electron-flow mechanism in chloroplasts and in photosynthetic bacteria is supported by recent evidence, presented in the succeeding article by M. Losada, F. R. Whatley and D. I. Arnon, that it is experimentally possible to replace water as the electron donor in non-cyclic photophosphorylation by chloroplasts and to demonstrate in this manner a bacterial type of non-cyclic electron flow in green plants: a reduction of pyridine nucleotide coupled with the oxidation of an exogenous electron donor and a simultaneous photophosphorylation but without evolution of oxygen.

These findings permit us to interpret with somewhat enhanced confidence, certain aspects of the biochemical evolution of photosynthesis which we have already discussed elsewhere[7,29].

The beginning of photosynthesis may be viewed as an emergence of a porphyrin that gave rise to chlorophyll and permitted the cell to use for metabolic purposes the energy of sunlight. In this primitive photosynthesis, the contribution of light-energy was probably limited to anaerobic cyclic photophosphorylation. No photochemically formed reductant would have been required for the photoassimilation of, say, acetate or for the assimilation of carbon dioxide so long as hydrogen gas was present in the atmosphere[29]. Oparin[54] and Miller and Urey[55] have summarized the evidence that in the early periods of the evolution of life the environment contained simple carbon compounds such as acetate and also hydrogen gas. Photosynthetic bacteria still show this primitive type of photosynthesis. *Chromatium*, for example, when supplied with hydrogen gas, uses it to reduce pyridine nucleotide in the dark, and seems then, as in the case of photoassimilation of acetate, to limit the use of light to the formation of adenosine triphosphate[29] (cf. ref. 30).

As hydrogen gas vanished from the primitive atmosphere, the photosynthetic cell probably became dependent on the photosynthetic apparatus for generating photochemically a strong reductant from such electron donors as succinate or thiosulphate. Light-energy now would have served a dual purpose: it would supply adenosine triphosphate by cyclic photophosphorylation and would also, by photooxidizing the electron donor, provide electrons for reducing pyridine nucleotides by a non-cyclic electron-flow mechanism. In organisms which contain, or can form adaptively, hydrogenase or nitrogenase (photosynthetic bacteria and algae), this phase of photosynthesis can be observed to-day as a photoproduction of molecular hydrogen or a photofixation of nitrogen gas.

The non-cyclic electron-flow mechanism in photosynthetic bacteria operates with such electron donors as reduced sulphur compounds or organic acids. Water (OH^-) became an electron donor only with the emergence of plant photosynthesis. As will be discussed in the following article, it now appears that this event was made possible because plants have evolved an auxiliary reaction dependent on light, in which the electrons from water (OH^-) are first raised to a potential similar to that of the electron donors in bacterial photosynthesis, and are then used in a second photochemical reaction—the non-cyclic electron flow common to plants and bacteria—in which pyridine nucleotide reduction is coupled with the formation of adenosine triphosphate.

Oxygen evolution is peculiar to green plants because it is the product of an auxiliary photochemical reaction, the oxidation of water (OH^-), that is found only in green plants. Water as an electron donor is therefore not involved in the two main events of non-cyclic photophosphorylation in plants and bacteria, the photoreduction of pyridine nucleotide and the coupled formation of adenosine triphosphate.

In both bacterial and plant photosynthesis the photosynthetic events proper are limited to the formation of adenosine triphosphate and reduced pyridine nucleotide by cyclic and non-cyclic photophosphorylation. This transformation of light into the common currency of cellular energy is fundamentally independent of carbon dioxide assimilation. There is no particular reason why adenosine triphosphate or a photochemically generated reductant could not be used for driving endorgonic cellular processes other than carbon dioxide assimilation.

In summary, the differences between bacterial and plant photosynthesis seem to centre on the electron donors that are consumed in the reduction of pyridine nucleotide. With some electron donors, as, for example, hydrogen gas, the reduction of pyridine nucleotide by photosynthetic bacteria is a dark reaction. With other electron donors, such as thiosulphate or succinate, an input of light-energy, in a non-cyclic electron-flow mechanism, becomes necessary for the oxidation of the electron donor and the reduction of pyridine nucleotide. Thus, in bacterial photosynthesis, the reduction of pyridine nucleotide may occur with or without an input of light energy depending on the particular electron donor used. By contrast, in photosynthesis of green plants, the reduction of pyridine nucleotide invariably requires an input of light energy because the physiological electron donor is water (OH^-).

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SEPARATION OF TWO LIGHT REACTIONS IN NONCYCLIC PHOTO-PHOSPHORYLATION OF GREEN PLANTS

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NONCYCLIC photo-phosphorylation[1-3] is a photochemical reaction in isolated chloroplasts which yields simultaneously three products: reduced pyridine nucleotide, adenosine triphosphate and oxygen, in accordance with reaction 1 (where P means orthophosphate):

$$ADP + P + TPN + H_2O \rightarrow ATP + TPNH_2 + \tfrac{1}{2} O_2 \quad (1)$$

Of the three products of reaction 1, only reduced pyridine nucleotide and adenosine triphosphate are required by chloroplasts in the conversion of carbon dioxide to carbohydrate[4-7]. Oxygen is an excreted by-product, incidental to the use of water (OH^-) as the electron donor in noncyclic photo-phosphorylation by chloroplasts[8]. Photosynthetic bacteria cannot use water as an electron donor, and here the key events of noncyclic photo-phosphorylation—the reduction of pyridine nucleotide and the formation of adenosine triphosphate—also occur, but are not accompanied by an evolution of oxygen (ref. 9 and unpublished work by Nozaki, M., Tagawa, K., and Arnon, D. I.).

Vernon and Zaugg[10], extending the earlier work of Vernon and Kamen[11], have shown that chloroplasts which have lost, through ageing or by poisoning, the capacity to evolve oxygen photochemically, are nevertheless able to reduce triphosphopyridine nucleotide photochemically with electrons donated by a reduced dye, 2,6-dichlorophenol indophenol. (In practice, the dye is supplied in catalytic amounts and kept in a reduced form by substrate amounts of added ascorbate.) Krasnovsky has found that chlorophyll solutions in pyridine can bring about a photochemical electron transfer from ascorbate or other electron donors to pyridine nucleotides or other electron acceptors[12].

We now wish to report that noncyclic photo-phosphorylation in chloroplasts has been experimentally separated into two distinct photochemical reactions: (*a*) a photo-oxidation of water (OH^-) that yields oxygen (reaction 2) and (*b*) a noncyclic photo-phosphorylation of the bacterial type, that is, a photo-reduction of triphosphopyridine nucleotide that is coupled with the formation of adenosine triphosphate (reaction 3). The two reactions have been separated by using reduced (A^-) and oxidized (A) indophenol dyes as summarized here:

$$H_2O + 2A \xrightarrow{\text{light}} \tfrac{1}{2} O_2 + 2A^- + 2H^+ \quad (2)$$

$$2A^- + TPN + 2H^+ + ADP + P \xrightarrow{\text{light}} TPNH_2 + ATP + 2A \quad (3)$$

$$\text{Sum: } TPN + ADP + P + H_2O \xrightarrow{\text{light}} ATP + TPNH_2 + \tfrac{1}{2} O_2 \quad (1)$$

Bacterial Type of Noncyclic Electron Flow in Chloroplasts

Table 1 shows that when catalytic amounts of 2,6-dichlorophenol indophenol[10] replaced water (OH^-) as the electron donor (treatment 3), illuminated chloroplasts formed adenosine triphosphate and reduced triphosphopyridine nucleotide without evolving oxygen. Similar results were obtained when reduced 2,3',6-trichlorophenol indophenol[13] was used instead of 2,6-dichlorophenol indophenol. The participation of water as an electron donor in these reactions was blocked by the addition of *p*-chlorophenyl dimethylurea[10] to, and the omission of chloride[14] from, the reaction mixture. When the participation of water as an electron donor was not blocked (Table 1, treatment 1), the reduction of triphosphopyridine nucleotide and the coupled phosphorylation were accompanied by evolution of

Table 1. Noncyclic Photo-phosphorylation of the Bacterial Type by Chloroplasts

No.	Addenda	Effective electron donor	Oxygen evolved (μatoms)	Triphosphopyridine nucleotide reduced (μmoles)	Adenosine triphosphate formed (μmoles)
1	None	Water	3·0	3·4	2·4
2	Chlorophenyl dimethyl urea	None	0	0·5	0·2
3	Dye, ascorbate, chlorophenyl dimethyl urea	Reduced dye (ascorbate)	0	3·2	3·4

Vessel 1 contained in a total volume of 3 ml.: washed chloroplast fragments (*P*1S2), prepared in the absence of chloride, containing 0·5 mgm. chlorophyll; 0·05 ml. purified spinach phosphopyridine nucleotide reductase, and the following, in micromoles: *tris*/acetate buffer, *p*H 8·0, 40; $MgSO_4$, 5; $K_2H^{32}PO_4$, 10; ADP, 10; TPN, 4; and KCl, 10. In vessel 2, KCl was omitted and 2×10^{-5} *M p*-chlorophenyl dimethylurea was added. Treatment 3 was the same as treatment 2 except that 20 μmoles ascorbate and 0·2 micromole dye (dichlorophenol indophenol) were added. The experiment was run for 20 min. at 15° (at a light intensity of 2,000 ft. candles). Oxygen was measured manometrically and triphosphopyridine nucleotide spectrophotometrically at 340 mμ.

The omission of chloride and the addition of chlorophenyl dimethyl urea, a powerful inhibitor of oxygen evolution, blocked the use of water as an electron donor in the chloroplast system. Catalytic amounts of dichlorophenol indophenol served as an electron carrier between ascorbate and the chloroplast system.

oxygen. When the participation of water was blocked but no substitute electron donor was supplied (treatment 2), no significant reaction occurred at all.

The 'bacterial' type of noncyclic electron-flow mechanism that is envisaged as operating in chloroplasts, when the participation of water as the electron donor is blocked (Table 1, treatment 3), is illustrated diagrammatically in Fig. 1. The electron donor, A^-, represents the reduced indophenol dye, and the electron acceptor, *B*, represents triphosphopyridine nucleotide (cf. reaction 3). In accordance with previous formulations, we envisage the formation of adenosine triphosphate as occurring during the electron transport between cytochromes and chlorophyll[15].

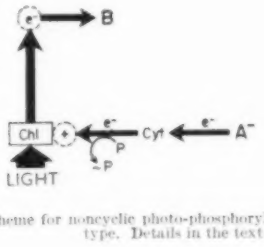

Fig. 1. Scheme for noncyclic photo-phosphorylation of the bacterial type. Details in the text

Photo-oxidation of Water as a Separate Reaction

In the experiments just described, catalytic amounts of phenol indophenol dye mediated the transfer of electrons from ascorbate to triphosphopyridine nucleotide. The dye was *reduced in the dark* by ascorbate and *oxidized in the light* by triphosphopyridine nucleotide. This electron transport was coupled with the formation of adenosine triphosphate (reaction 3).

We will now describe experiments in which catalytic amounts of phenol indophenol dye mediated the transfer of electrons from water (OH^-) to ferricyanide. The dye was *reduced in the light* by water (OH^-) (resulting in an evolution of oxygen) and was *oxidized in the dark* by ferricyanide. This photo-oxidation of water did not result in the formation of adenosine triphosphate (reaction 2).

Indophenol dyes have long been known to be reduced photochemically by chloroplasts[13,16] in a Hill reaction; but unlike such Hill reagents as ferricyanide[1–3], the photo-reduction of 2,3′,6-trichlorophenol indophenol is not linked with photo-phosphorylation[13,17]. Krogmann and Jagendorf[13] drew from this "the inference . . . that reduction of indophenol dye is primarily by means of uncoupled electron transport", possibly by a "'short circuit' of that part of the electron transport system which is coupled to phosphorylation".

We have examined the effect of catalytic amounts of indophenol dyes on a variant of noncyclic photo-phosphorylation in chloroplasts where ferricyanide replaces triphosphopyridine nucleotide (reaction 4).

$$2Fe^{3+}Cy + H_2O + ADP + P \xrightarrow{\text{light}} 2Fe^{2+}Cy + \tfrac{1}{2}O_2 + ATP + 2H^+ \quad (4)$$

The results are shown in Table 2. The addition of catalytic amounts of indophenol dye to the ferricyanide system has almost completely suppressed the formation of adenosine triphosphate without impairing oxygen evolution (treatments 2 and 4). The addition of catalytic amounts of indophenol dye has thus converted the noncyclic photo-phosphorylation reaction with ferricyanide into a more limited reaction, a photo-oxidation of water that is not accompanied by photo-phosphorylation.

Table 2. Separation of Photo-production of Oxygen and Photo-phosphorylation by Chloroplasts

No.	Treatment	Effective electron acceptor	Oxygen produced (μatoms)	Adenosine triphosphate formed (μmoles)
1	Ferricyanide	Ferricyanide	4·5	4·7
2	Ferricyanide Dye *A* +	Oxidized dye (Ferricyanide)	5·1	1·3
3	Ferricyanide	Ferricyanide	3·3	3·5
4	Ferricyanide Dye *B* +	Oxidized dye (Ferricyanide)	3·5	0·8

The reaction mixture contained, in a final volume of 3 ml., unwashed chloroplast fragments containing 0·2 mgm. chlorophyll, and the following in micromoles: *tris*/HCl buffer, *p*H 8, 100; $MgSO_4$, 5; $K_2H^{32}PO_4$, 10; adenosine diphosphate, 10; potassium ferricyanide, 10. Dye *A* (0·2 μmole 2,6-dichlorophenol indophenol) or dye *B* (0·05 μmole 2,3′,6-trichlorophenol indophenol) was added where indicated. 10 per cent potassium hydroxide was present in the centre well of the manometer vessels. The reaction was run for 12 min. at 15° C. in the light (2,000 ft. candles). The gas phase was nitrogen.

In the experiments illustrated in Table 2, it was possible to measure the oxygen evolution that accompanies the photo-reduction of indophenol dyes because they were supplied in catalytic amounts and were kept in the oxidized state by the presence of substrate amounts of ferricyanide. A direct measurement of oxygen evolution that accompanies the photo-reduction of 2,6-dichlorophenol—or 2,3′,6-trichlorophenol indophenol—is difficult to make because these dyes, when used in concentrations that are required for manometric measurements of evolved oxygen, absorb too much light. This reduces the effective light intensity to such an extent that chloroplasts cease to evolve oxygen[16]. Indophenol dyes are therefore generally used as Hill reagents in low concentrations, and their photo-reduction is measured not manometrically by oxygen evolution but optically, by their decolorization[16,18].

The observed effect of indophenol dyes could not be attributed to inhibiting, uncoupling, 'short-

circuiting' or by-passing a phosphorylation step in reaction 4. As shown in Table 3, the addition of indophenol dyes to the noncyclic photo-phosphorylation system with triphosphopyridine nucleotide (reaction 1) did not suppress photo-phosphorylation and has, in fact, slightly increased it. As in the ferricyanide system (Table 2), the addition of the dye to the triphosphopyridine nucleotide system did not affect the oxygen evolution.

Table 3. Effect of Indophenol Dyes on Noncyclic Photo-phosphorylation by Chloroplasts in the Presence of Triphosphopyridine Nucleotide (TPN)

No.	Treatment	Effective electron acceptor	Oxygen produced (μatoms)	Adenosine triphosphate formed (μmoles)
1	TPN	TPN	3·3	3·2
2	TPN Dye A +	TPN	3·1	4·2
3	TPN	TPN	3·0	3·3
4	TPN Dye B +	TPN	3·1	3·6

Experimental conditions were the same as for Table 2, except that the ferricyanide was omitted. 4 μmoles TPN and 0·1 ml. of a purified preparation of triphosphopyridine nucleotide reductase from spinach were added in each treatment.

The photo-oxidation of water is diagrammatically represented in Fig. 2, where A stands for oxidized dye. The reaction requires chloride. We regard the photo-oxidation of water by chloroplasts as an auxiliary reaction to supply electrons at an elevated reducing potential for a second photochemical reaction, the bacterial type of noncyclic photo-phosphorylation, during which a phosphorylation, coupled with the reduction of pyridine nucleotide, actually occurs (Fig. 1).

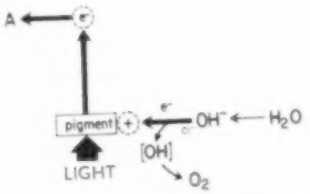

Fig. 2. Scheme for photo-oxidation of water by chloroplasts. Details in the text

It appears that noncyclic photo-phosphorylation in green plants combines the two component photochemical reactions: (*a*) the auxiliary reaction for photo-oxidation of water (Fig. 2) and (*b*) the bacterial type of noncyclic photo-phosphorylation (Fig. 1) that is used both by photosynthetic bacteria and by green plants for photo-reduction of pyridine nucleotide and a coupled phosphorylation.

Fig. 3 depicts the combined scheme for noncyclic photo-phosphorylation in green plants that we now envisage. B is the electron acceptor, triphosphopyridine nucleotide under physiological conditions, but it can be replaced in cell-free systems by ferricyanide. Under special conditions, molecular oxygen can also serve as the electron acceptor B in cell-free systems, and this would result in an "oxidative photosynthetic phosphorylation"[17] (compare Fig. 1) or "oxygen-catalysed cyclic photo-phosphorylation"[18] (compare Fig. 3). In the latter case an oxygen exchange reaction would result. The consumption of oxygen at one end (B in Fig. 3) would balance the production of oxygen at the other (bottom of Fig. 3).

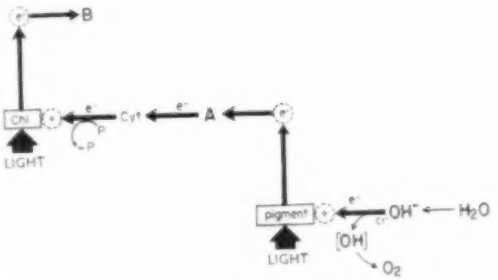

Fig. 3. Scheme for noncyclic photo-phosphorylation in green plants. Details in the text

The intermediate A in Fig. 3 is both the electron acceptor for the first, and the electron donor for the second, light reaction (compare Figs. 1 and 2). We have used indophenol dye as an experimental device to separate the two photochemical reactions. The natural intermediates with which the dyes have interacted have not been identified. They may include cytochromes or a quinone[20].

According to the scheme shown in Fig. 3, electron acceptors in chloroplast reactions may be divided into those which react in position B and those which react in position A. Only the first group, exemplified by triphosphopyridine nucleotide, would give a coupled photo-phosphorylation. 2,3′,6-Trichloro-phenol indophenol[12,17] is an example of the second group. However, certain compounds, although reacting preferentially in position A, may also be capable of reacting in position B.

Since indophenol dyes are reduced by illuminated chloroplasts much more rapidly than ferricyanide[12,21], a catalytic amount of the dye can convert noncyclic photo-phosphorylation with ferricyanide into a photo-oxidation of water reaction (Table 2). In the absence of the dye, ferricyanide acts as a terminal electron acceptor B in noncyclic photo-phosphorylation, and oxygen evolution is accompanied by formation of adenosine triphosphate (cf. Fig. 3 and Table 2, treatments 1 and 3). When the dye is present, it is repeatedly reduced during the photo-oxidation of water and re-oxidized by ferricyanide in the dark (cf. Fig. 2 and Table 2, treatments 2 and 4). Here oxygen is produced without the formation of adenosine triphosphate (Fig. 2). However, indophenol dyes do not convert noncyclic photophosphorylation with triphosphopyridine nucleotide into a photo-oxidation of water reaction (Table 3). The dark re-oxidation of the reduced dye by triphosphopyridine nucleotide is precluded by the difference in the respective normal redox potentials (0·217 V. for 2,6-dichlorophenol indophenol and −0·320 V. for triphosphopyridine nucleotide).

Triphosphopyridine Nucleotide Reductase from *Chromatium*

The separation in chloroplasts of the photo-phosphorylation reaction from the photo-oxidation of water underlines the similarities and the differences between noncyclic electron-flow mechanisms in photosynthetic bacteria and in green plants. The difference is most pronounced at the electron donor end, where green plants, but not photosynthetic bacteria, can use water (OH^-) as the electron donor.

Unlike photosynthetic bacteria, chloroplasts contain neither nitrogen-fixing enzymes nor hydrogenase.

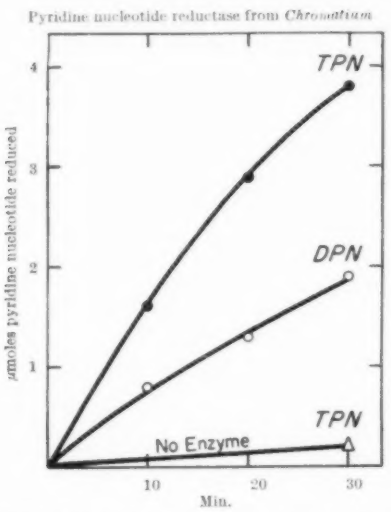

Fig. 4. Pyridine nucleotide reductase from *Chromatium*. Experimental conditions as in Table 4, except that $K_2H^{32}PO_4$ and ADP were omitted. 4 μmoles DPN or TPN were added as indicated; ...

As a consequence, the electron acceptor end of the noncyclic electron-flow mechanism in chloroplasts can be coupled neither to photofixation of nitrogen nor to photoproduction of hydrogen gas, but only to reduction of pyridine nucleotide. However, certain species of green and blue-green algæ constitute exceptions among plants. They can fix nitrogen photosynthetically or they can become adapted to using hydrogen gas as a reductant in photosynthesis (literature cited in ref. 9).

Despite the differences in enzymic composition that govern nitrogen fixation and hydrogen utilization, the noncyclic electron-flow mechanisms in bacteria and chloroplasts are basically similar at the electron acceptor end. This view is strengthened by the recent isolation of a photosynthetic pyridine nucleotide reductase from *Chromatium* and *R. rubrum*. As shown in Fig. 4, the bacterial enzyme catalysed the photochemical reduction of pyridine nucleotides by chloroplasts that have been deprived of their own pyridine nucleotide reductase. When tested by adding to washed chloroplasts, the bacterial pyridine nucleotide reductase was similar to the chloroplast pyridine nucleotide reductase in reducing preferentially triphosphopyridine nucleotide over diphosphopyridine nucleotide[22,23].

The reduction of triphosphopyridine nucleotide was coupled with oxygen evolution when the bacterial enzyme was added to a chloroplast preparation that by itself could not reduce triphosphopyridine nucleotide and thereby evolve oxygen (Table 4). These findings again support the conclusion that reduction of triphosphopyridine nucleotide and oxygen evolution are basically separate phenomena. The bacterial pyridine nucleotide reductase cannot bring about a coupling of pyridine nucleotide reduction with oxygen evolution in a bacterial system.

Table 4. Photochemical Oxygen Evolution catalysed by Pyridine Nucleotide Reductase from *Chromatium*

Min.	Oxygen evolved (μatoms)	Triphosphopyridine nucleotide reduced (μmoles)
5	0·75	0·76
10	1·60	1·40
20	2·78	2·39
30	3·97	3·18

The reaction mixture contained, in a final volume of 3 ml.: washed chloroplast fragments containing 0·3 mgm. chlorophyll; and the following in micromoles: *tris* buffer, *p*H 7·8, 100; $MgCl_2$, 5; ADP, 10; $K_2H^{32}PO_4$, 10; TPN, 6; and a purified pyridine nucleotide reductase preparation from *Chromatium*. The reaction was run at 15° in the light.

Concluding Remarks

The concept of photosynthesis presented now is a further elaboration, in the light of new experimental findings, of that presented earlier[15,9]. It is now possible to envisage, in more detail, the electron transfer reactions from water (OH^-) that result in oxygen evolution. We had first suggested that this may be a dark reaction between water (OH^-) and a cytochrome component peculiar to green plants[15]. Such a reaction, although it appeared thermodynamically possible[15], could not be described in terms of the properties of known cytochrome constituents of green cells[24]. We next suggested that the interaction between OH^- and cytochromes may require an additional input of energy[19], and now it appears that this energy is supplied by an auxiliary light reaction. This auxiliary light reaction is found in green plants but not in photosynthetic bacteria, and explains why photosynthesis is accompanied by evolution of oxygen only in green plants.

The separation of photo-oxidation of water from the photo-phosphorylation reaction is consistent with the view that photosynthesis in green plants involves the participation of more than one light reaction. Warburg *et al.*[25] found in *Chlorella* an enhancement of photosynthesis under red light by the addition of catalytic amounts of blue light and attributed this effect to a photoactivation of a photosynthetic enzyme ('Luminoferment'). The maximum effectiveness of the catalytic light was at 460 mμ. Another light effect, which seems more directly related to our results, was reported by Emerson *et al.*[26], who observed that enhancement of photosynthesis in *Chlorella* under red light was produced by light of shorter wave-length; in their experiments the supplementary light was not catalytic but of an intensity "sufficient to give measurable photosynthesis". Emerson[27] found the maximum effectiveness of the supplementary light in *Chlorella* at 480 mμ, and a lower peak of effectiveness at about 655 mμ, coinciding approximately with the maximum absorption peaks of chlorophyll *b*.

Emerson[27] suggested from these and similar observations with other algæ that, at the long wavelengths of light absorbed only by chlorophyll *a*, full efficiency of photosynthesis depends on the absorption of light of shorter wave-length by an accessory photosynthetic pigment: chlorophyll *b* in *Chlorella*, or phycobilins in other algæ. The supplementary light in this view is not used as a catalyst but is consumed as a substrate.

The enhancement effect of supplementary light has been confirmed by other investigators[28–30]. Of special relevance to our concept is the conclusion of Myers and French[28] that their results favour "the idea that the interaction [of the light absorbed by chlorophyll *a* and *b*] takes place between chemical products of two separate reactions initiated by light" and the report of Govindjee *et al.*[30] that the enhancement effect is associated with the oxygen-liberating

mechanism rather than with reduction of carbon dioxide.

That two light reactions, with the participation of water, are involved in the photosynthetic cytochrome system of both green plants and photosynthetic bacteria has been postulated by Hill and Bendall[24]. However, our findings support the conclusion that green plants possess a special photochemical reaction, photo-oxidation of water, that is absent in photosynthetic bacteria. We have not identified the 'pigment' (Fig. 2) that is involved in the photo-oxidation of water. This pigment is more likely to be chlorophyll *b* or an accessory pigment that is found only in higher plants and algae, rather than chlorophyll *a* (or the closely related bacteriochlorophyll) found in all photosynthetic cells*.

* *Note added in proof.* Additional evidence that oxygen evolution in chloroplasts is catalysed by a photosynthetic pigment different from that involved in the photoreduction of triphosphopyridine nucleotide and photophosphorylation was recently obtained in this Laboratory in experiments with monochromatic light. In the red region of the spectrum, the photo-oxidation of water was most effective around 644 mμ (a maximum absorption peak for chlorophyll *b*), whereas the effectiveness of red light for photoreduction of triphosphopyridine nucleotide and photophosphorylation showed markedly different spectral characteristics (cf. Arnon, D. I., "Photosynthetic Phosphorylation and a Unified Concept of Photosynthesis", Photosynthesis Symp., Fifth Int. Congr. Biochem., Moscow, 1961).

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INTRACELLULAR STRUCTURES IN TUMOURS INDUCED BY POLYOMA

By LEON DMOCHOWSKI, CLIFFORD E. GREY, ELIZABETH BERECZKY* and JULIAN BLICHARSKI†

Section of Virology and Electron Microscopy, University of Texas M. D. Anderson Hospital and Tumour Institute; Department of Microbiology, Baylor University College of Medicine, Texas Medical Center, Houston, Texas

CERTAIN animal viruses have been described as forming crystals in the nucleus or cytoplasm of the infected cells (for literature, see ref. 1). Light microscopy of histochemically stained specimens and electron microscopy of similar specimens have indicated the nature and significance of these structures in virus development. The tumour-inducing virus, polyoma, has also been observed to form crystalline-like arrangements of characteristic particles (270 Å.) in the nuclei of mouse embryo cells and of kidneys of hamsters infected with this agent[2]. These particles, however, could not be found in the cells of tumours induced by polyoma in the kidneys of hamsters[3]. An electron microscope study of kidneys of mice, rats, and hamsters infected by polyoma has revealed characteristic particles (270 Å.) in the nuclei of cells, two or three weeks following infection with the virus[3]. Fewer particles have been found in the cells of proliferative lesions in the kidneys of infected animals of the three species, and even smaller numbers in the cells of tumours induced by polyoma in the kidneys of rats and hamsters[4].

In this article the results of extensive electron microscope studies of tumours induced in the kidneys of mice, rats (obtained through the courtesy of Dr. Bernice E. Eddy) and hamsters by tissue culture preparations of polyoma virus will be presented. Only material from kidneys showing gross nodular tumours confirmed by histological diagnosis has been used in the present work. Tissue specimens were fixed in 1 per cent osmium tetroxide buffered with veronal-acetate containing 1 per cent sucrose, dehydrated in ethyl alcohol of graded dilutions, embedded in *n*-butyl methacrylate, and sectioned with glass knives. To enhance the contrast, the sections mounted on grids were floated on a saturated solution of uranyl acetate with the specimen facing down for 30 min.–24 hr. Alternate

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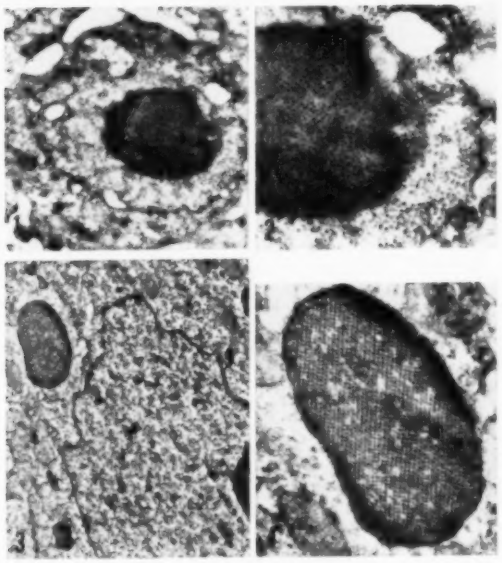

Fig. 1. A cell in polyoma-induced kidney tumour of a hamster with characteristic particles scattered in the nucleus and an inclusion body. (× 10,000)
Fig. 2. Part of Fig. 1 at higher magnification, showing the particles in the inclusion body of the nucleus. (× 21,000)
Fig. 3. Inclusion in the cytoplasm of a cell of a hamster kidney tumour. (× 5,000)
Fig. 4. Cytoplasmic inclusion showing a regular array of particles. (× 22,500)

thin (300 Å.) and thick sections (2μ) were cut. The thick sections were studied by the light microscope, following staining with the Feulgen method. The thin sections were examined in an RCA Type *EMU 3E* electron microscope.

A number of non-specific changes, such as shrinkage of the cells, pyknosis of the nucleus, condensation and vacuolization of the cytoplasm, and alterations in the structure of mitochondria have been observed in the cells of kidney tumours of mice, rats and hamsters. Changes which could be regarded as specific have also been observed in some of the cells in scattered areas of kidney tumours of animals of all three species. Particles, similar to those already described in polyoma-infected cells *in vivo*[5] and *in vitro*[5], have been found scattered throughout the nucleus as shown in Fig. 1, and with more detail in Fig. 2. Vacuolization of the nucleus may also be found (Fig. 1) and the nuclear membrane may be replaced by a tightly packed row of particles. The same nucleus may contain an inclusion body (Fig. 1) filled with characteristic particles in a regular arrangement, as shown at higher magnification in Fig. 2. Occasionally, cells are encountered with enlarged nuclei containing particles scattered at random (Fig. 3) and with cytoplasm showing mitochondria in various stages of degeneration and inclusions, varying from 0·5μ to more than 2μ (Fig. 3). Some of the inclusions are filled with the characteristic particles arranged in regular pattern, as in the largest inclusion shown in the top left-hand corner of Fig. 3, and at higher magnification in Fig. 4. In spite of crystalline-like arrangement of the particles, the structure must not be considered as a crystal until examined by methods such as electron diffraction. In the corresponding thicker (2μ) sections of the same specimen examined by the light microscope, the intranuclear inclusions containing the characteristic particles are positive to Feulgen staining. Cells of tissue cultures of polyoma-infected mouse embryo, examined in the light microscope at various intervals of time after infection, have shown the gradual formation of Feulgen-positive inclusions in the nuclei followed by the appearance of similarly staining inclusions in the cytoplasm[6]. On examination in the electron microscope, the characteristic particles have been found within these inclusions[6]. These inclusions stain as DNA, following acridine orange treatment[7], and the stained inclusions are not removed by digestion with DNA-ase[6]. However, pre-treatment of the cells with pepsin, followed by digestion with DNA-ase, removes the inclusions[6]. This is further indication that the inclusions are of viral nature, as shown in tissues infected by other viruses[7].

Cells have also been observed with most of the cytoplasm filled with strongly osmiophilic material (Fig. 5) in which numerous regular arrays of particles may be seen among mitochondria and vacuoles (Fig. 6). Measurements have been made of 20 particles which appear elliptical in section (Fig. 6). They measure an average of 330 Å. in length and 260 Å. in width. The average distance from centre to centre of the particles is 390 Å. Assuming the particles in the section to be distorted spheres, the diameter of the equivalent circular particles is calculated as 290 Å. In view of the possibility that a peripheral zone of the particles has not been made visible by osmium tetroxide and uranyl acetate, the diameter of the particles, based on the measured centre-to-centre distance and the equivalent circular diameter of the particle, is approximately 390 Å. This measurement of the particle size approximates to that reported for polyoma particles in dried and shadow-cast preparations from infected tissue cultures (440 Å., ref. 8), and in concentrated or partially purified suspensions stained by the negative-contrast method (453 Å.) with potassium phosphotungstate[9].

The particles, which at lower magnifications appear uniformly dense without an apparent membrane or capsid[9], appear to have an internal structure in stained sections examined at higher magnifications (Fig. 7). Similar structure has been seen in the particles present in the sections of cells of infected mouse embryos stained with uranyl nitrate[10]. The structure has been interpreted as reticular and as

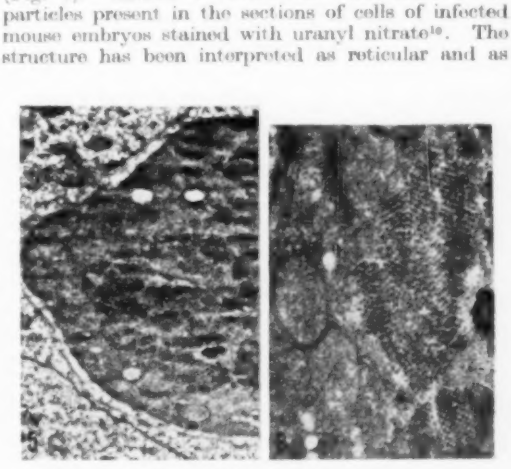

Fig. 5. Cytoplasm of hamster kidney tumour cell showing mitochondria and regular formations of particles. (× 5,000)
Fig. 6. Part of Fig. 5, showing three mitochondria and the characteristic particles. (× 21,000)

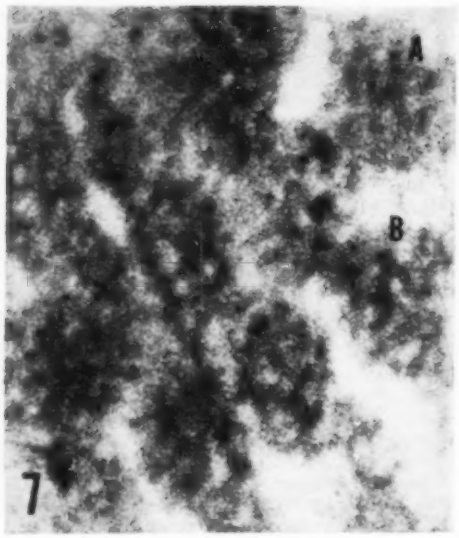

Fig. 7. Some of the particles with outlines of internal structure. (× 540,000)

containing an outer coat of lower contrast[10]. A comparison of the appearance of some of the particles (Figs. 7 and 8) with that of the particles stained with potassium phosphotungstate[9] reveals a certain similarity in structure. A detailed description of the structure of the particles present in sections of the kidney tumours will be published.

In conclusion, it may be stated that the characteristic particles in polyoma-infected tissue culture cells and in the kidney cells of infected mice, rats and hamsters are also present in smaller numbers in the cells of kidney tumours of animals of the three species. The observation of particles of similar size and shape in different types of cells infected with

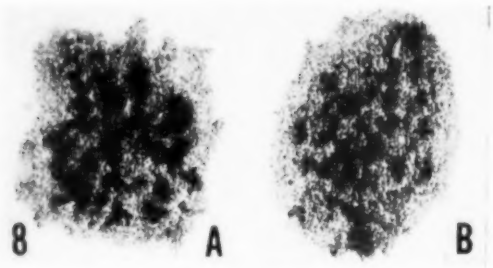

Fig. 8. Schematic drawing of the appearance of two particles shown in Fig. 7

polyoma and in tumours induced by this virus in different species gives further support to the identification of these particles as polyoma virus particles.

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AN ELECTRON MICROSCOPE STUDY OF THE 'CHLORIDE-SECRETORY CELL' OF *SALMO SALAR* L., WITH REFERENCE TO PLASMA-ELECTROLYTE REGULATION

By Dr. LAWRENCE T. THREADGOLD
Department of Zoology, Queen's University, Belfast, Northern Ireland
AND
Dr. ARTHUR HILLIER HOUSTON
Department of Zoology, University of Manitoba, Winnipeg, Canada.

THE teleostean kidney is unable to secrete a hypertonic urine. Because of this, the marine teleost must rely on extra-renal electrolyte excretory processes to reduce the salt load incurred in drinking sea-water to replace body-water lost by the process of osmotic dehydration. Smith[1] suggested that the gills might be the site of the extra-renal excretory mechanism, while Keys[2], through the use of a heart–gill preparation, was able to demonstrate the ability of the gill of the eel to transport chloride against a concentration gradient. Afterwards, Keys and Willmer[3] described a type of cell in the gill of this and other species which they suggested might be the site of an active transport system, and tentatively termed it the 'chloride-secretory cell'.

Although similar cells have been described in a variety of marine, euryhaline and anadromous teleosts[4–10,14,15] under varying experimental conditions, their function in electrolyte excretion has not as yet been firmly established. Nevertheless, the adaptive responses observed after transfer between fresh and salt water[5,6], histochemical demonstrations of high cellular chloride content in fish adapted to sea-water[5], correlations between ability to tolerate

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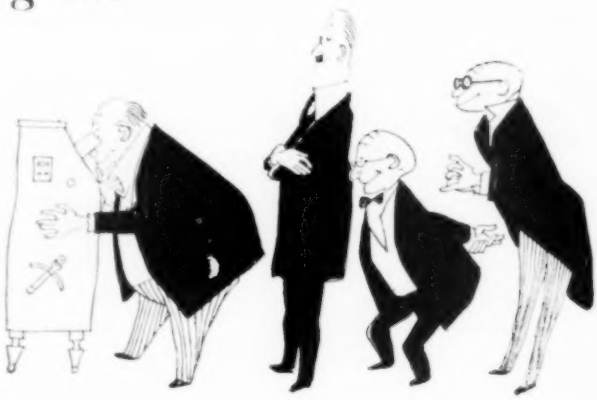

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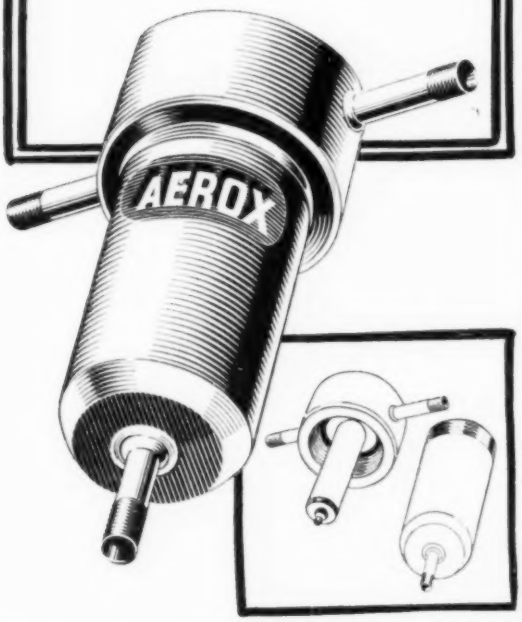

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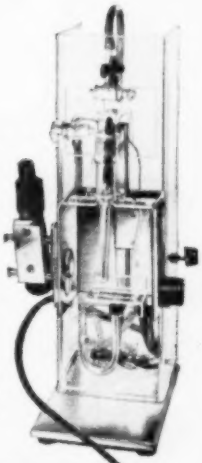

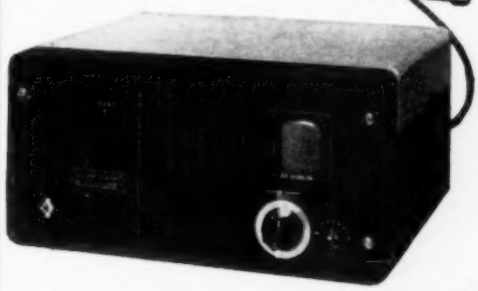

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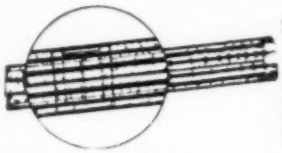

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Hair

Admonui, ne trux caper iret in alas,
Neve forent duris aspera crura pilis
(Ovid, Ars Amatoria, Book iii, 193)

In its own way, and in the right place, hair is a thing of beauty, but scientifically it takes its place with wool, nail, hoof, beak, etc., as just one manifestation of keratin, a fibrous protein which exists in coiled (α) form and straightened (β) form, and exhibits unique elastic qualities.

Physiologically, as Wells has put it, a hair is a rod of dead cells proliferated by a living root. It is the result of a metabolism having as its end-product a coloured, self-lubricated, preferably curled fibre.

The colouring material is melanin, and the lubricant is sebum. Because hair is an end-product, and because our ultimate interest in it is evocative, we can change its appearance and heighten its appeal without prejudice to its biological growth.

But whereas our knowledge of the structure of keratin is well-founded—due to the work of Astbury and Speakman in Leeds, Pauling in the U.S., and the C.S.I.R.O. team in Australia—and we have a reasonably clear picture of it in terms of stereochemical configuration, disulphide linkage and hydrogen bonding, the picture is not so clear when we think of keratin in terms of the cortex and cuticle of hair and the differences they exhibit in comparison with wool. Nor is the precise significance of sebum understood as a lubricant of the hair.

At Unilever Research at Isleworth, therefore, Holmes and his colleagues are carrying out a programme of research on hair—its structure, its colour, its lubrication, and the influence of extraneous materials on them—to obtain a greater understanding of it.

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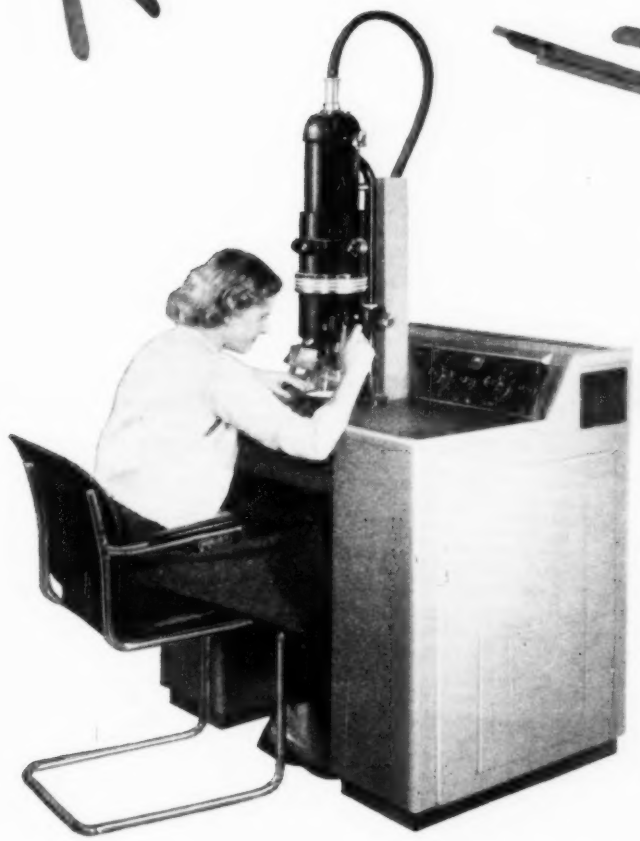

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The place of the Particle Accelerator in Basic Research

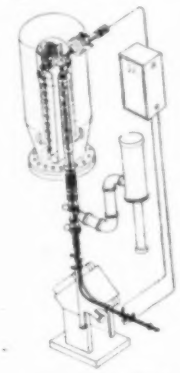
Atomic Displacement by High-energy Particles-VI

Research on the interaction of high-energy particles with atoms in the solid phase has received new impetus with the availability of controlled beams of electrons, positive ions, and monoenergetic neutrons from Van de Graaff accelerators. Investigations in this important field of solid-state physics and chemistry have been the subject of several recent surveys and books. [1, 2, 3].

Determination of Displacement Threshold

Of great interest has been the determination of the threshold for displacement in semi-conductors, by measurements that can be made with electrons of several hundred Kev. Increasing the energy above the threshold value causes the atom to recoil with sufficiently high kinetic energy to cause further displacements in collisions with neighbouring atoms. Electrons cannot transfer large amounts of energy to these recoil atoms because of the large mass difference, but changes can be effected by heavy particles if they have sufficient kinetic energy. Although high-energy protons and deutrons have been used for solid-state studies, these particles do not provide the best results because they lose energy to the lattice electrons and, at low energies, have a limited range.

Neutrons can be produced with a Van de Graaff accelerator by bombardment of targets such as Li^7 and H^3 with protons or deutrons. The resulting neutron beam is monoenergetic, with a high flux, making it ideal for displacement studies. It does not suffer from the broad energy spectrum of pile-produced neutrons and has relatively low gamma background.



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Thermal Spike

In general the monoenergetic neutrons produced by particle accelerators have energies above the displacement threshold. Their interaction with the atomic nuclei can be described by classical hard-sphere collisions and the subsequent recoil atom energy is sufficient to produce electronic excitation of neighbouring atoms. This phenomenon, originally described in 1923[4], is generally known as a thermal or displacement spike. It is one of the most difficult and yet most interesting phenomena in solid-state radiation reactions.

Seitz, in describing the theory of the high-temperature spike[2], has made an estimate of the volume affected and of the temperature and pressure produced in this region by transfer of incident energy to a lattice atom. Using assumptions deduced from rather meager experimental evidence, it can be shown that the affected volume has a radius approximately ten times the atomic radius. In this volume, the temperature is above the melting points of most solids and the pressure due to the high local temperature is of the order of 10^7 dynes per $cm.^2$ Although these conditions last for only about 10^{-11} seconds, which is brief in comparison with most chemical reactions, they could be useful for certain selected systems.

One of the exciting possibilities of such local thermal spikes is the use of the extremely high temperatures and pressures for chemical solid-state reactions. Little has been done in this field because of the difficulty of analyzing the results. With the availability of low-energy monoenergetic neutrons from a Van de Graaff, without the complication of Coulomb encounters with the lattice atoms, there appears hope that this promising field will attract more attention.

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sea-water and the occurrence of these cells[6,7], and finally the close correlation between their ontogenetical appearance in the migratory salmonids[8–10] and the development of tolerance to sea-water in these species[10–12] provide strong indirect evidence favouring the hypothesis.

On the other hand, several authors have felt that the 'chloride-cell' was not, in fact, associated with electrolyte metabolism. Thus Bevelander[13], on the basis of extensive comparative studies, concluded that the only specialized cells in the branchial epithelium were mucous cells. However, as Copeland[5] has pointed out, at least some of the cell types described by Bevelander as being 'mucous-secretory' had most of the cytological characteristics commonly attributed to 'chloride cells'.

Burden[14] has disclosed a puzzling situation in the course of work on the osmoregulatory ability of hypophysectomized *Fundulus*. Death in fresh-water following the destruction of the pituitary was accompanied by asthenia, massive decreases in levels of plasma chloride and by atrophy of branchial mucous cells. No obvious variations in 'chloride cell' structure were observed. Copeland[5] has suggested that such cells in *Fundulus* may have dual polarity, excreting electrolytes in sea-water, and functioning in an absorptive role in fresh-water. The situation described by Burden may possibly be accounted for as a failure of the electrolyte absorptive process to cope with increased outward diffusion of electrolytes through surfaces no longer protected by a mucous layer.

Finally, Parry, Holliday and Blaxter[15] have objected to the secretory hypothesis on the grounds that a similar type of cell is found in the pseudobranch in large numbers, where by virtue of blood supply and position, a secretory function was not considered likely; that bilaterally pseudobranchectomized animals were able to survive in sea-water over long periods of time, and that under such conditions no adaptive increases in branchial 'chloride cell' numbers occurred.

In view of the conflicting evidence on this theoretically important aspect of teleostean osmoregulation, we decided to include an electron microscope examination of the branchial epithelium in an investigation of the processes of parr–smolt transformation and adaptation to sea-water in hatchery-reared yearling Atlantic salmon. A part of this work has already been published in *Nature*[16].

Gill structure was examined in parr adapted to fresh-water, and for purposes of comparison in a two-year-old trout (*Salvelinus fontinalis*), in smolt held for 72 hr. in sea-water (400–420 m.moles chloride/litre), and in smolt acclimatized to sea-water for 10 days. Reference to Figs. 1 and 2 of ref. 16 indicates that under these conditions parr displayed some loss of regulatory ability associated with the early stages of parr–smolt transformation, smolt adapted for 72 hr. to sea-water exhibited maximum deviations from the chloride-levels of fresh-water plasma, while fish held in sea-water for 10 days were considered to have completed the process of adaptation.

Gill filaments from the first gill arch were fixed in Dalton's fixative at *p*H 7·4 and embedded in methacrylate. Sections were cut on a Porter–Blum microtome with a diamond knife and photographed at × 2,000 to × 8,000 on a Phillips *E.M.* 75.

Salvelinus fontinalis (Fig. 1). In this species the 'chloride-secretory cells' were usually columnar, with basally situated ovoid nuclei. There were numerous mitochondria, the majority of which lay between the nucleus and the free pole of the cell. A relatively small number lay basal to the nucleus. Few mitochondria were present in the zone immediately below the apical plasma membrane. Individual mitochondria tended to be elongate with their long axes parallel to that of the cell.

In the mitochondria-free apical zone there were numerous dense micro-vesicles, some of which were either in contact with mitochondria or with the plasma membrane.

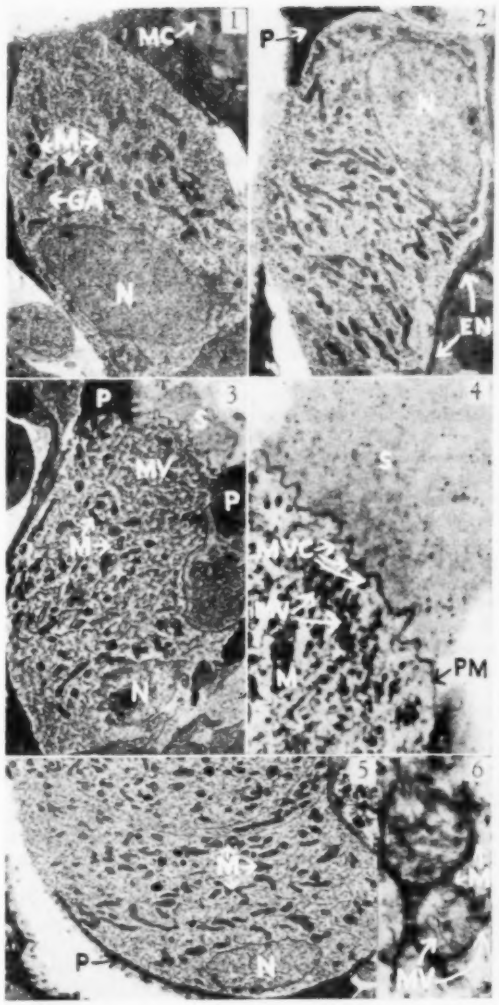

Fig. 1. *Salvelinus fontinalis*. *M.*, mitochondria; *G.A.*, Golgi apparatus; *N.*, nucleus; *M.C.*, mucous cell. (× 3,750)

Fig. 2. *Salmo salar*. Parr. *E.N.*, endothelium of blood vessel. *P.*, platelet cell. (× 4,500)

Fig. 3. *S. salar*. Smolt. 72 hr. adapted. *S.*, secretory vesicle; *M.V.*, micro-vesicles. (× 3,375)

Fig. 4. *S. salar*. Smolt, 72 hr. adapted. *P.M.*, plasma membrane; *M.V.C.*, micro-vesicles contacting the plasma membrane and discharging. (× 9,000)

Fig. 5. *S. salar*. Smolt. 10-day adapted. (× 3,000)

Fig. 6. *S. salar*. Smolt. 72 hr. adapted. Mitochondria with associated micro-vesicles. (× 45,000)

A variable number of juxta-nuclear Golgi bodies and short lengths of endoplasmic reticulum were also present in the cytoplasm.

The apical plasma membrane was overlain to a variable extent by adjacent electron-dense platelet cells, but in fully developed secretory cells the free membrane was usually the full width of the cell and bulged outward beyond the outer limits of the platelet cells. The membrane of the chloride cell was characterized by the presence of minor undulations, whereas that of the platelet cell usually projected, forming a number of microvilli. External to the cell membrane, there was in most instances a narrow amorphous layer, presumably a secretion product. This suggested a low level of secretory activity in these cells.

Salmo salar (Fig. 2). The 'chloride cells' of the Atlantic salmon parr were in general similar to those described for the trout. Rectangular rather than columnar cells were numerous. Minor differences were found in: (1) a more open cytoplasm with fewer mitochondria, (2) crenated nuclei, and (3) the presence of pale bodies resembling greatly swollen mitochondria. The cells gave the appearance of secretory exhaustion in some instances.

Salmo salar, 72-hr. smolt (Figs. 3, 4 and 6). In the 72-hr. smolt, the 'chloride cells' were either columnar or rectangular. In several features they were clearly distinct from those seen in parr. Mitochondria were often extensively vacuolated and had irregularly arranged cristæ. In some instances the mitochondria had ruptured and tubular cristæ were in the process of being shed into the cytoplasm (Fig. 6).

There was marked evidence of secretory activity with many cells having a mass of amorphous material containing scattered dense granules lying immediately beyond the free apical border. Such cells usually lay in a 'pit' formed by the adjacent platelet cells (Fig. 3).

In cells characterized by a high degree of secretory activity, the apical region was crowded with micro-vesicles of variable size resembling the cristæ shed from the more central mitochondria. With the production of a large secretory mass, the apical region lacked micro-vesicles and the mitochondria-free zone was larger (Fig. 4).

The 'chloride cells' of the 72-hr. smolt were, then, characterized by intense secretory activity of the merocrine type, and apparently related to mitochondrial activity.

Salmo salar, 10-day smolt (Fig. 5). In contrast to the cells just described, those of the 10-day smolt resembled the 'chloride cells' of the parr and trout. They varied from columnar to rectangular in shape, and contained numerous, clearly delineated mitochondria. Few apical vesicles were present and the free surface of the apical cell membrane was reduced. The mass of the secretory product was much less than that seen in the 72-hr. fish, and in many instances no secretory product could be distinguished. In general, a picture of reduced activity characterized the 'chloride cell' of the 10-day smolt.

The 'chloride cell' as described here is comparable to that demonstrated in earlier cytological studies[4–10,14,15] in distribution, overall shape and relative size, in the occurrence of a predominantly basal nucleus, high mitochondrial content, and in the presence of a mitochondria-free apical zone. We have therefore concluded that the type of cell described here represents that investigated in the investigations already cited, and as such is clearly distinct from the typical mucous cell.

The great similarity of these cells in the fresh-water adapted trout, in the parr salmon and in the 10-day smolt is strikingly correlated with chloride-levels in the plasma, which in each instance were apparently regulated within specific limits. It is perhaps notable that among this group some signs of secretory exhaustion were apparent in the cells of the parr, as evidence of an electrolyte imbalance is seen in these fish during the early stages of parr–smolt transformation[16].

In the occurrence of vacuolated mitochondria and an apical zone crowded with micro-vesicles, and in the presence of a large extracellular secretory mass, the cells of the 72-hr. smolt differed markedly from those encountered in animals in dynamic equilibrium with their medium. Such cells gave every indication of heightened secretory activity, which was correlated with sharp increases in electrolyte-levels in the body fluids. The process of secretion appeared to involve formation of large numbers of micro-vesicles by, or in close association with, mitochondria. These vesicles fused with the plasma membrane and were discharged, or released, to form the extracellular secretory mass.

This formation of micro-vesicles in close association with the apical mitochondria and seemingly related to the formation of the extracellular secretory mass is of particular interest in view of Copeland's demonstration by histochemical techniques of a marked concentration of chloride in this area in *Fundulus* adapted to sea-water[5]. It seems reasonable to conclude, then, that the extracellular mass described here is at least in part composed of inorganic electrolytes, possibly excreted in an organic matrix.

The present work corroborates earlier conclusions regarding adaptive changes resulting from transfer of fish from fresh-water into sea-water. Evidence for a secretory function has been presented. In view of Copeland's work already cited[5] and the correlation of enhanced secretory activity and structural changes with maximum increases in electrolyte-levels in plasma, we concluded that this work provides additional support for the hypothesis that the branchial 'chloride-secretory cell' is associated with extra-renal electrolyte excretion in teleosts adapted to the marine environment.

We acknowledge the financial support of the National Research Council of Canada and the Nova Scotia Research Foundation. A full report of this investigation, carried out in the Department of Anatomy and Zoological Laboratory, Dalhousie University, Halifax, Canada, will be published elsewhere.

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LETTERS TO THE EDITORS

SPACE SCIENCE

Satellite Orbit Perturbations in Vector Form

THE first-order perturbation theory of satellite orbits can be formulated simply and elegantly in terms of constants of the unperturbed motion using vector methods. For let the equation of motion be:

$$\ddot{\mathbf{r}} = -\frac{\mu}{r^3}\mathbf{r} + \mathbf{F} \qquad (1)$$

where $\mu = GM$ (M = mass of the earth), and $\mathbf{F}$ is a general perturbing force (per unit mass). The energy E and angular momentum $\mathbf{h}$ (both per unit mass) defined as:

$$E = \frac{1}{2}\dot{\mathbf{r}}^2 - \frac{\mu}{r}\,; \quad \mathbf{h} = \mathbf{r} \times \dot{\mathbf{r}} \qquad (2)$$

are constants of the unperturbed motion. Hamilton's integral, which may be written in the form[1]:

$$\mathbf{k} = \frac{\mu}{r}\mathbf{r} + \mathbf{h} \times \dot{\mathbf{r}} \qquad (3)$$

provides a further constant of the unperturbed motion under an inverse square law force. Taking scalar products with $\mathbf{h}$ and $\mathbf{r}$, $\mathbf{k}$ is seen to be a vector of length μe lying along the major axis in the direction of apogee. The orbit is over-determined by E, $\mathbf{h}$ and $\mathbf{k}$ which give seven scalar equations.

In the presence of the perturbing force, E, $\mathbf{h}$ and $\mathbf{k}$ are no longer constant, but from (1) satisfy:

$$\begin{aligned} \dot{E} &= \dot{\mathbf{r}} \cdot \mathbf{F} \\ \dot{\mathbf{h}} &= \mathbf{r} \times \mathbf{F} \\ \dot{\mathbf{k}} &= (\mathbf{h} \times \mathbf{F}) + (\mathbf{r} \times \mathbf{F}) \times \dot{\mathbf{r}} \end{aligned} \qquad (4)$$

When $\mathbf{r}$, $\dot{\mathbf{r}}$ and $\mathbf{h}$ are taken to refer to the unperturbed orbit, these are the equations of first order in the perturbing force.

It is best to discard two components of $\dot{\mathbf{k}}$ since this has the most complicated equation. Then E gives the change in the semi-major axis ($E = -\mu/2a$), $\mathbf{h}$ the rotation of the plane of the orbit and the change in eccentricity [$h^2 = \mu a(1-e^2)$], while the rotation of the major axis in the plane is given by the component of $\dot{\mathbf{k}}$ parallel to the minor axis.

The quantities E, $\mathbf{h}$ and $\mathbf{k}$ will give the osculating elements at any time. However, for most purposes the interesting effects are those of period longer than the satellite in its orbit. These are given by the mean rates of change obtained by integrating (4) over a complete orbit, using either the true or the eccentric anomaly, as appropriate, as the independent variable.

The results can often be expressed partly in vector form. For the simple case of a constant force $\mathbf{F}$, corresponding, for example, to radiation pressure on a uniform sphere the orbit of which is entirely in sunlight, the effects are given by:

$$\dot{E} = 0, \; \dot{\mathbf{h}} = \frac{3}{2}\frac{a}{\mu}\mathbf{k} \times \mathbf{F}, \; \dot{\mathbf{k}} = \frac{3}{2}\mathbf{h} \times \mathbf{F} \qquad (5)$$

This method has been applied to solar and lunar perturbations and to the effect of radiation pressure when part of the orbit is in darkness. It has also been applied to the second harmonic term in the Earth's gravitational field giving results in agreement with previous work[2].

A treatment essentially in terms of E, $\mathbf{h}$ and $\mathbf{k}$ appears to have been proposed first by Herrick[3], and Musen[4] has applied it to the case of a constant perturbing force giving effectively the results of (5) above.

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The Cosmic-Ray Observations of December 4, 1957

A RECENT communication from Legrand and Helary[1] reports an increase in the total component of cosmic radiation at Paris on December 4, 1957. The increase amounted to about 18 per cent and extended over three hours with the maximum occurring at 2245 U.T. A small increase in the nucleonic component at Thule preceded the Paris event by about three hours. Optical and radio data indicate a flare of importance 1 on the western hemisphere of the Sun at 1657 U.T. (ref. 2).

Intensity enhancements at the surface of the Earth are comparatively rare, only eleven cases having been observed. However, these cosmic ray flare events appear to have certain well-defined properties in common. These include correlation in time with a visible or inferred solar flare, a steep integral rigidity spectrum and approximate simultaneity of occurrence at all points on the Earth's surface accessible to the radiation. The steep rigidity spectrum is indicated by the relative rarity of events seen in ground-level neutron monitors as compared to detectors flown in balloons and the small magnitude (or absence) of the increases measured in the meson component compared to the nucleonic enhancements.

Collins and Jelly[2] have reported that ionospheric conditions were normal throughout the period in which the increase in cosmic radiation occurred and have also pointed out that solar activity was low. Since an undisturbed ionosphere at the time of a cosmic-ray flare is an unlikely event, it was thought worth while to examine the intensity of cosmic radiation monitored at the Sulphur Mountain Laboratory at this time. The Laboratory is on a mountain near Banff, Alberta, Canada (lat. 51·2° N., long. 115·6° W.), at 7,500 ft., and houses both neutron and meson monitors. The neutron monitor, consisting of three boron trifluoride counters in a paraffin – lead pile, has a counting-rate of 100,000/hr., and a cubical Geiger counter telescope containing 10 cm. lead

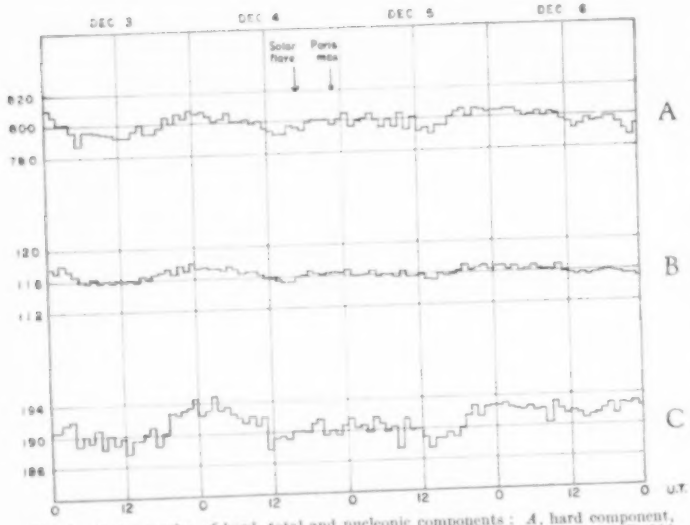

Fig. 1. Rates per hr. of hard, total and nucleonic components: *A*, hard component, *S.F.*, 1,024; *B*, total ionizing component, *S.F.*, 2,048; *C*, nucleonic component, *S.F.*, 512

measures the intensities of the total ionizing component and the meson component. The counting-rates are respectively 240,000 and 80,000/hr.

The results, corrected for pressure, are plotted in Fig. 1 for the period December 3–6, 1957. It is clear that all three components show no unusual variations during the period of the reported event; this result is in agreement with other collected data[1] from a large number of stations. Since the data from Paris and Thule are not supported in the records of neighbouring stations, and since the times of the two maxima are well separated from each other and from the time of the optical flare, the events reported are difficult to reconcile with the emerging pattern of the development of cosmic-ray flares. In the absence of corroborative evidence of cosmic ray enhancements or of subsequent Forbush decreases at other stations, and with the negative results of the ionosonde data, it appears reasonable to assume that the reported increase was not associated with an injection of solar protons.

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Cosmic Noise Measurements from 1960 τ_1 at 3·8 Mc./s.

We wish to report the results of an experiment to measure cosmic noise at 3·8 Mc./s. from an Earth satellite.

Solid-state radiometers, for the measurement of cosmic noise above the ionosphere, have been designed and built at the Defence Research Board Telecommunications Establishment in Ottawa. A single frequency unit, of a type already described[1], was part of the 1960 τ_1 (*Transit* 1*A*) payload and operated satisfactorily for the life of the battery pack.

The satellite telemetry transmitter was turned on and off via a command link, and since command decisions were dependent on the payload battery condition, on some passes there was no telemetry. For the period June 24–27, 1960, the orbital parameters for τ_1 were as follows: inclination angle (i) = 66·7°, apogee altitude, 1,070 km.; perigee altitude, 625 km. The perigee and apogee occurred in the southern and northern hemispheres, respectively. Telemetry stations were located at Washington, D.C.; Fort Myers, Florida; Antigua, British West Indies; Santiago, Chile; Woomera, Australia; and Ottawa, Canada.

The equipment aboard *Transit* 1*A* consisted of a superheterodyne receiver with a 40 kc./s. pass-band centred on 3·8 Mc./s., and to reduce errors arising from changes in gain and band-width, the receiver was switched continuously between the antenna and a resistive reference noise source. Temperature tests from −30° C. to +30° C. showed no measurable change in receiver sensitivity. The absolute value of the sensitivity was limited by the efficiency of the antenna since the main payload requirements did not permit the optimum antenna geometry and weight.

For the purposes of calibration, a single-turn terminated loop was permanently attached to each antenna. The loop was calibrated for a known field-strength, and was then used to obtain the input–output characteristics of the radiometer. Antenna pattern measurements indicated negligible departure from that of the ideal magnetic dipole.

For a terminated antenna in thermal equilibrium, the sky temperature is given by:

$$T = \frac{PA}{KB} \qquad (1)$$

where A = effective antenna aperture in m.^{2}; P = power flux in watts m.$^{-2}$ c.p.s.$^{-1}$; K = Boltzmann's constant; T = temperature in deg. K.; B = band-width c.p.s.; also

$$P = \frac{E^2}{Z} \qquad (2)$$

where Z = the impedance of the medium in ohms; E = electric field strength of the incident energy in μV./m. for a noise band-width of 40 kc./s.

Table 1 is given in terms of the equivalent linearly polarized free-space field-strength, for a noise band-width of 40 kc./s. in a plane perpendicular to the major axes of the antennæ. To determine P, a correction factor of 1·5 must be made to the value of field strength, E_m, given in Table 1. This correction assumes an omni-directional noise field, whereas the calibrating field was linearly polarized. The expression for P must therefore be rewritten as:

$$P = 1{\cdot}5\,E_m^2/Z \qquad (3)$$

Where the results were not affected by the radiometer sensitivity, the accuracy was dependent on antenna calibration and the stability of the telemetry link. At low signal-levels, the accuracy was limited by the noise signals generated in other apparatus

Table 1. CENTRE FREQUENCY 3·5 Mc./s., NOISE BAND-WIDTH 40 Kc./s.

June 24, 1960				June 25, 1960				June 26, 1960				June 27, 1960			
Time (G.M.T.)	Field strength (μV./m.)	Height (km.)	Comments	Time (G.M.T.)	Field strength (μV./m.)	Height (km.)	Comments	Time (G.M.T.)	Field strength (μV./m.)	Height (km.)	Comments	Time (G.M.T.)	Field strength (μV./m.)	Height (km.)	Comments
Blossom Point, Maryland, U.S.A. and Ottawa, Canada															
2010	0·8	1,055	Good record	0321	0·8	920	Good record	0303	0·65	880	Earth breakthrough, excellent record	0248	0·6	845	Good record
0338	0·8	880	Good record	0504	0·65	860	Good record	0448	0·8	860	Earth breakthrough, excellent record	0431	0·6	880	Good record, breakthrough
1827	0·7	1,045	Good record												
Fort Myers, Florida, U.S.A.															
2005	0·8	1,035	Good record	No records				No records				0247 0433	0·6 0·6	830 810	Good record Breakthrough
Santiago, Chile															
No records				No records				No records				0535	0·85	640	Poor record
Woomera, Australia															
0433	1·50	890	Fair record	1718 0412 1535	1·95 1·46 2·04	640 790 655	Fair record Poor record Good record	0409	1·50	890	Good record	No records			
Pantig (Antigua) British West Indies															
No records				1805	0·90	1,030	Poor record	0310	0·80	780	Poor record	0255 0438	0·65 0·65	780 790	Poor record Breakthrough

within the satellite and to a lesser extent by the noise figure of the receiver. From considerations of these factors it is estimated that, for all stations except Woomera, the probable error was +1, −5 db., and for Woomera, the probable error was ±1 db.

Certain features of the results are worthy of further discussion. On the night of June 25, 1960, high-level fluctuating signals were recorded when the satellite was approximately 45° N. latitude in the vicinity of Ottawa, Canada, moving on a north-to-south pass. The fluctuations occurred at the spin-rate of the satellite, which had its spin axis inclined at an angle of approximately 20° to an Earth tangent. At the time of the pass, the f_0F2 at Ottawa, Canada, was 3·5 Mc./s. and it is believed that the fluctuations were due to radiation from the Earth reaching the radiometer. This phenomenon occurred on two successive passes.

Records taken in the northern hemisphere (Ottawa, Canada and Blossom Pt., Maryland) showed no variation in noise power between day and night, whereas those taken at Woomera, Australia, showed considerable variation. Table 1 shows that the local night readings at Woomera were about 2·5 db. higher than the local day readings which were, in turn, 3 db. higher than those for the northern hemisphere.

The one usable record from Santiago, Chile, does not agree with the Woomera recordings even though both stations are at approximately the same latitude. The quality of the Santiago recordings is poor, due to telemetry station difficulties, whereas the Woomera recordings are very good. No ground ionospheric data, which might have resolved this discrepancy, are as yet available from either location.

The support and co-operation of the Applied Physics Laboratory of the Johns Hopkins University are gratefully acknowledged. Thanks are also due to the National Aeronautics and Space Administration of the United States for recording telemetry data and to the Naval Ordnance Test Station of the U.S. Navy for supplying payload battery power. The contributions of Dr. D. F. Page, of the Defence Research Telecommunications Establishment, in the receiver design and of E. D. R. Shearman, of the Department of Scientific and Industrial Research, Slough, England, in the interpretation of the data are also acknowledged.

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GEOPHYSICS

Odd Harmonics of the Earth's Gravitational Field

VARIATIONS in the perigee radius have been observed for three satellites so far. The first of these is *Vanguard I* (1958 beta), which was used by O'Keefe *et al.*[1] in calculating the third and fifth axial harmonics of the Earth's field. The other two are *Transit* IB (1960 gamma) and *Transit* IIA (1960 eta). Nominal values of the pertinent orbit parameters of these three satellites are listed in Table 1. The data

Table 1. Orbital Parameters

Satellite	a, Earth radii	e	i, radians	C_p
Vanguard I	1·3615	0·1903	0·5977	−0·00102
Transit 1B	1·0871	0·0278	0·8949	−0·00115
Transit 1IA	1·1312	0·0309	1·1639	−0·00026

on *Vanguard* are from O'Keefe *et al.*[1], those on *Transit* 1B are given by Guier[2], and those on *Transit* IIA have not been published before.

For our purposes, it is most convenient to express the perigee radius as a function of θ_p, the latitude of perigee, according to:

$$r_p = r_{p_0} + C_p \sin \theta_p \qquad (1)$$

Values of C_p are listed in Table 1. The probable errors in the values of C_p are believed to be about 7×10^{-5} for *Vanguard* and about 6×10^{-5} for the two *Transits*.

The dependence of C_p upon the orbit parameters is given by:

$$C_p = (J_3/2J_2) - (5J_5/4J_2P^2)\left(1 + \frac{3}{4}e^2\right)\left[\left(1 - \frac{7}{2}f + \frac{21}{8}f^2\right)/\left(1 - \frac{5}{4}f\right)\right] + (35J_7/16J_2P^4)\left(1 + \frac{5}{2}e^2 + \frac{5}{8}e^4\right)\left[\left(1 - \frac{27}{4}f + \frac{99}{8}f^2 - \frac{429}{64}f^3\right) \div \left(1 - \frac{5}{4}f\right)\right] \qquad (2)$$

In this, $f = \sin^2 i$ and $P = a(1 - e^2)$. The terms involving J_3 and J_5 were apparently first derived by O'Keefe *et al.*[1]. The convention that we are using in defining the coefficients J_2, J_3, etc., is specified by writing the gravitational potential as:

$$U = (1/r) - \sum_{n=2}^{\infty} (J_n P_n(\sin\theta)/r^n + 1) \qquad (3)$$

where $P_n(x)$ is the nth Legendre polynomial, normalized to equal unity when $x = 1$. The value of J_2 is approximately $1{\cdot}083 \times 10^{-3}$.

Substituting the data from Table 1, and the estimated errors, into equation 2 yields:

$$J_3 = (-2{\cdot}36 \pm 0{\cdot}14) \times 10^{-6}$$
$$J_5 = (-0{\cdot}19 \pm 0{\cdot}10) \times 10^{-6}$$
$$J_7 = (-0{\cdot}28 \pm 0{\cdot}11) \times 10^{-6}$$

These should be compared with the values:

$$J_3 = (-2{\cdot}4 \pm 0{\cdot}3) \times 10^{-6}$$
$$J_5 = (-0{\cdot}1 \pm 0{\cdot}1) \times 10^{-6}$$

that O'Keefe *et al.* obtained from *Vanguard* I data alone, neglecting J_7.

It is interesting that the value of J_7 obtained here is larger than the value of J_5, although the difference is less than the probable error. It may be that the values of the harmonics beginning with the fifth will be determined principally by the topography, and will thus tend to decrease slowly but erratically with increasing order.

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Measurements of Air Radioactivity in Italy and their Relation to the First Sahara Atomic Explosion

On the occasion of the French atomic explosion of February 13, 1960, in the Sahara, a control system of radioactivity in the air was set up covering the whole of Italy. The controlling stations of the system were equipped to collect samples of atmospheric dust by means of suction through a filter. The β-activity of the samples was measured 120 hr. after they had been collected.

The stations were established throughout Italy (from north to south) as follows, the heights being in metres:

	Place	Height above sea-level (m.)		Place	Height above sea-level (m.)
1	Pian Rosà	3,488	15	Bari	4
2	Pallanza	212	16	Napoli	2
3	Ispra	251	17	Resina	610
4	Trieste	8	18	Brindisi	8
5	Milano	123	19	Alghero	21
6	Bologna	64	20	Monte Scuro	1,716
7	Genova	364	21	Elmas (Cagliari)	18
8	Monte Cimone	2,173	22	Messina	54
9	Pisa	3	23	Palermo	30
10	Livorno	3	24	Chinisia (Trapani)	74
11	M. Terminillo	1,875	25	Pantelleria	185
12	Casaccia (Rome)	154	26	Cozzo Spadaro (Syracuse)	40
13	Vigna di Valle	270			
14	M. S. Angelo	844			

Two intervals showing increased activity were observed which, although very weak, may be considered as a consequence of the passage of the tropospheric atomic dust cloud originated by the explosion of February 13, 1960, after it had rounded the Earth following an eastward course along the parallels of latitude. The first interval occurred between February 27 and March 2; the second interval between March 7 and March 15.

The two intervals were probably due to the passage of the radioactive dust cloud after it had completed the first passage around the Earth, divided in two parts driven along different itineraries by winds at different altitudes.

In the first interval, higher values were registered, all corresponding to the stations located at greater heights:

Place	Height above sea-level (m.)	Counts
Pian Rosà	3,488	15·00 pc./m.³
Monte Cimone	2,173	8·00 ,,
M. Terminillo	1,875	0·75 ,,
M. Scuro	1,716	1·8 ,,
M. S. Angelo	844	0·6 ,,
Resina	610	0·22 ,,
Vigna di Valle	270	0·8 ,,
Livorno	3	0·15 ,,

During the second interval, stations located on mountains were particularly involved by the passage of the radioactive cloud. In fact, measurements of radioactivity gave the following values:

Place	Height above sea-level (m.)	Counts
Pian Rosà	3,488	2·20 pc./m.³
Monte Cimone	2,173	1·0 ,,
M. Terminillo	1,875	0·5 ,,
M. Scuro	1,716	3·4 ,,
M. S. Angelo	844	0·6 ,,
Vigna di Valle	270	0·24 ,,
Livorno	3	0·12 ,,

The highest activity observed at these stations during the second half of March 1960 was:

Place	Counts
Pian Rosà	0·4 pc./m.³
Monte Cimone	0·2 „
M. Terminillo	0·2 „
M. Scuro	0·2 „
M. S. Angelo	0·2 „
Vigna di Valle	0·2 „
Livorno	0·09 „

The activities registered were always markedly lower than the highest recorded in previous years. In 1958 Naples and Pisa registered the following highest values, namely, 67 pc./m.³ and 43 pc./m.³ respectively; in 1959 the highest value at Pian Rosà was 22·5 pc./m.³.

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PHYSICS

Spin-Lattice Relaxation of Pairs of Chromium Ions in Ruby

The electron paramagnetic resonance spectrum of ruby contains many weak lines as well as those of single chromium ions. It has been established from the dependence of their intensity on the chromium concentration that most of these arise from pairs of ions. An example of the 'pair' spectrum is given in Fig. 1. A complete analysis is not available, but Rimai *et al.* have identified the lines due to nearest-neighbour pairs, and find an antiferromagnetic exchange interaction $JS_1.S_2$, with $J = 250$ cm.$^{-1}$, to be present'. These pairs are not observed at low temperatures, being then almost all in their singlet ground-state. Most of the lines, however, come from more widely separated ions, and many persist even at 1·3° K. The variation of their intensities with temperature indicates that levels separated from the ground-state by 0–15 cm.$^{-1}$ are present.

The spin-lattice relaxation of the ion-pairs in a crystal containing 0·12 per cent chromium has been measured from the recovery after saturation of some of the more prominent lines in the X-band 'pair' spectrum. The recovery was usually adequately described by a single time-constant. The measured times are shown in Figs. 2(*A*–*E*). In part of the range the times vary more or less inversely as the temperature T, suggesting relaxation through single phonons with energies less than kT. At higher temperatures the relaxation times vary more rapidly, presumably as multiple-phonon processes assume importance. At the lowest temperatures the relaxation of some of the lines becomes independent of temperature. This may be evidence that phonons with energies equivalent to several deg. K. are concerned in the relaxation: a

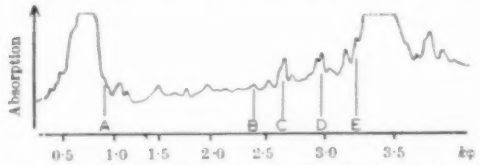

Fig. 1. Paramagnetic resonance of chromium pairs in ruby (room temperature spectrum, radio-frequency 9,400 Mc./s., magnetic field parallel to crystal axis). Single-ion lines occur at fields of 750 and 3,350 φ

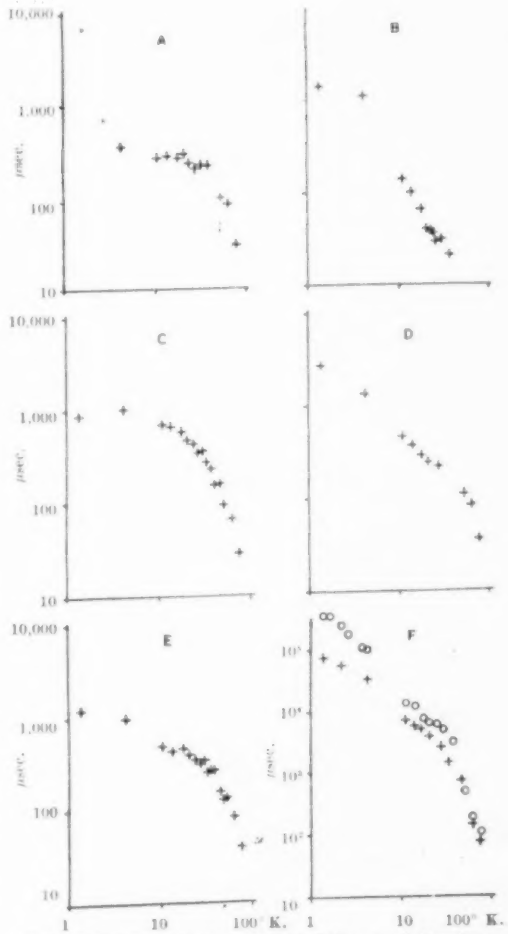

Fig. 2. Relaxation in a ruby containing 0·12 per cent chromium. *A*–*E*, Relaxation times measured from 'pair' lines *A*–*E* in Fig. 1; *F*, relaxation times of 750-φ line (circles) and of 3350-φ line (crosses) in single-ion spectrum

variation with $\{1-\exp(-E/kT)\}$ is to be expected in the relaxation between two excited levels if this takes place through single phonon processes to and from a third level the energy of which is lower by an amount E. Relaxation via an upper level, with times varying as $\{\exp(E/kT)-1\}$, has been observed in cerium[3]. Further experiments are necessary to decide between this explanation and the alternative that at the lowest temperatures the measurements were merely of the rate of transfer of spin energy from the pairs to the single ions by temperature-independent spin-spin relaxation.

The relaxation of two of the single-ion lines has also been examined. The relaxation times were measured from the last part of the recovery, to reduce the effects of spin-spin relaxation[3]. The measured times are shown in Fig. 2*F*.

The relaxation of the pairs is 10–300 times faster than that of the single ions in the same crystal. Fast relaxation is to be expected in pairs of ions with spins greater than $\frac{1}{2}$ if strong exchange interactions are present, as transitions involving energies of the order of the exchange energy are possible[3]. Since this

energy may be much greater than that of the single-ion transitions, far more lattice modes may be available to bring about the relaxation of a pair than of a single ion. Possible means whereby the lattice vibrations might induce the transitions include the 'Van Vleck' modulation of the electric fields of diamagnetic neighbours, and also modulation of anisotropic exchange interactions, or of isotropic interactions in the presence of some other anisotropy.

The relaxation of the pairs seems sufficiently rapid to account for much of the relaxation of the single ions. For example, let us assume that the pairs relax on the average 100 times more rapidly than the single ions, and that each single-ion line is effectively coupled by spin-spin processes to ten 'pair' lines. Then if the populations of the 'pair' levels are 0·1 per cent of those of the single-ion levels, we find that the energy reaching the lattice from the pairs accounts for the entire loss from the single ions. This may possibly provide an explanation of the concentration-dependent relaxation times commonly observed in ruby[4].

A more detailed account of this work will be published on its completion. I should like to thank Dr. R. J. Elliott and Prof. J. H. Van Vleck for helpful discussions, and Drs. C. B. P. Finn, R. Orbach and W. P. Wolf for letting me have details of their work in advance of publication.

Note added in proof. Since this was written, an analysis of the 'pair' spectrum has been reported (Weber, M. J., Rimai, L., Statz, H., and de Mars, G., *Bull. Amer. Phys. Soc.*, **6**, 141; 1961). The authors present indirect evidence for fast 'pair' relaxation, and point out the possible connexion with concentration-dependent single-ion times.

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'Memory' of Initial Remanent Magnetization and Number of Repeating of Heat Treatments in Low-temperature Behaviour of Hæmatite

It has recently been reported that the remanent magnetization of hæmatite (αFe_2O_3) indicates a marked thermal hysteresis phenomenon during a cooling–heating cycle to interpose the transition temperature of about 250° K., where the rotation of spin axis occurs: on cooling below 250° K. and then re-heating to room temperature, the direction of the remanent magnetization is still memorized in the specimen, although its intensity does not recover its original value before the cooling–heating cycle[1]. A satisfactory explanation of such behaviour has not so far been given, though much work has been carried out on this problem[2,3]. In this communication, new information on the 'memory' phenomenon will be reported.

The thermo-remanent magnetization and the isothermal remanent magnetization of the synthetic hæmatite powders were subjected to the repeated cooling–heating cycles between liquid nitrogen temperature and room temperature. In both cases,

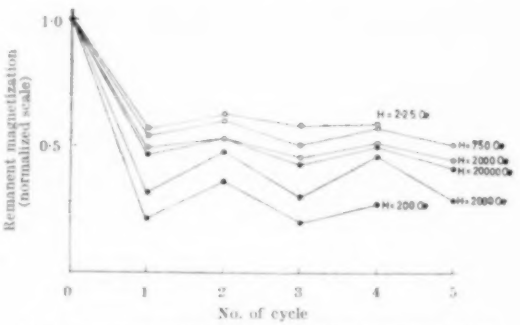

Fig. 1. Remanent magnetization of synthetic hæmatite powders at room temperature after the repeated cooling–heating cycles. ○, Thermo-remanent magnetization; ●, isothermal remanent magnetization

change in the intensity of remanent magnetization after the temperature cycling was measured by an astatic magnetometer at the room temperature. Recoveries relative to the initial remanent magnetizations, which were acquired in various magnetic fields, after the repeated cooling–heating cycles, are shown in Fig. 1 in reference to the numbers of cycle, where the thermo-remanent magnetization is indicated by circles and the isothermal remanent magnetization by dots.

It is found in the figure that the recovery of the intensity of remanent magnetizations after the first cycle is closely dependent on the process leading to the remanent magnetization. The rate of recovery is generally large in the case of thermo-remanent magnetization compared with isothermal remanent magnetization. It is also found that in the case of thermo-remanent magnetization the larger the applied magnetic field, the smaller the rate of recovery, while in the case of isothermal remanent magnetization the relation between the rate of recovery and the applied field is reversed. In the very strong field, the rate of recovery in both cases is confused.

Another interesting fact obtained in this work is that the intensity of remanent magnetization changes systematically in response to the number of cycles for the repeated temperature cycling. As seen in Fig. 1, the marked decrease in the first cycle is followed by a 'zig-zag' change for the repeated cycling, in a series of experiments the intensity of remanent magnetization corresponding to any even number of cycles ($2n$th cycle) being generally larger than that corresponding to the consecutive odd number of cycles ($2n \pm 1$ th cycle). This 'zig-zag' phenomenon is also dependent on the process leading to the remanent magnetization. A relative amplitude of the 'zig-zag' to the initial remanent magnetization is largest in the isothermal remanent magnetization produced by a weak magnetic field and smallest in the thermo-remanent magnetization produced by a weak magnetic field. The amplitude amounts to about 15 per cent of the initial remanent magnetization in the former case, and only a few per cent in the latter case. It is also confirmed from a similar experiment shown in Fig. 2 that this 'zig-zag' phenomenon is conserved for a large number of cooling–heating cycles, though the amplitude decreases almost exponentially.

Considering that a similar phenomenon is observed in the hæmatite powder synthesized from iron (Johnson Matthey and Co., Ltd.), which is believed to be pure, this 'zig-zag' phenomenon may not be attribut-

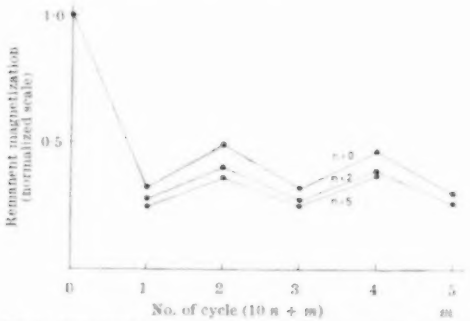

Fig. 2. Systematic change in the isothermal remanent magnetization of synthetic hæmatite powders at room temperature with large numbers of cooling–heating cycles (H = 2,000 oersteds)

able to any special characteristic of the hæmatite specimen. It has been confirmed, too, that the spatial orientation of the hæmatite specimen, relative to the geomagnetic field during the cooling–heating cycles, has no appreciable effect on the 'zig-zag' phenomenon. In view of these results, it may be concluded that this 'zig-zag' phenomenon is essentially related to a parasitic ferromagnetism of hæmatite.

A unified mechanism, through which the combined phenomena, namely, the memory and the 'zig-zag' of the remanent magnetization of hæmatite at low transition temperature can be interpreted simultaneously, may be required in future work.

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The Use of the Term 'Flux'

Recent suggestions[1] for the adoption of a unit of neutron intensity in reactor physics and technology provide an opportunity to raise the issue of non-ambiguity. In the physical sciences, the term 'flux density' has been used since the earliest days to denote units of the relevant quantity per unit area traversed normally per unit time. The term 'flux', then, describes the product 'flux density × area'. Apparently owing to the isolation of nuclear physics during the period 1939–45, the practice seems to have become established in certain branches of this subject and its associated technologies of using the term 'flux' as a short hand for 'flux density'. This ambiguous use tends to deprive both terms of their original significance, and has led to considerable confusion.

The ambiguity is quite unnecessary and its continuance is surely most undesirable. We should like to propose, therefore, that any unit such as the chad, whatever its magnitude, should be defined as a unit of flux density, its dimensions being $NL^{-2}T^{-1}$.

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CRYSTALLOGRAPHY

Structure of Vapour-deposited Carbon

Although considerable information is available in the published literature on the effect of temperature and pressure of deposition on the microstructure and density of vapour-deposited carbon, only little has been reported on the pyrolytic graphite formed under these conditions from the crystallographic point of view. The purpose of this communication is to present X-ray results obtained in these laboratories from analysis of a series of experimental deposits produced over a range of high temperatures at a pressure of 50 mm. mercury. The perfection of three-dimensional crystallinity exhibited in some specimens produced at moderate deposition temperatures at this pressure is, to our knowledge, a new observation.

Pyrolytic graphite is deposited on a substrate by passing a vapour source of carbonaceous material at reduced pressure and raised temperature over a heated surface. The pyrolytic graphite produced in this way exhibits stacking such that the atomic basal planes are parallel, as in natural graphite, but the atomic layers are randomly oriented to each other. This arrangement destroys the periodic repetition of atom positions in the c axial direction, and produces greatly different electrical and thermal conductivities in the directions normal and parallel to the substrate surface.

X-ray patterns of pyrolytic graphite specimens produced in these laboratories, as well as a commercial graphite and a commercial pyrolytic graphite sample, were obtained with a 114·59-mm. Debye–Scherrer powder camera using copper K_α radiation. The details of the X-ray analyses of these samples are shown in Table 1 and Fig. 1. These results indicate that the

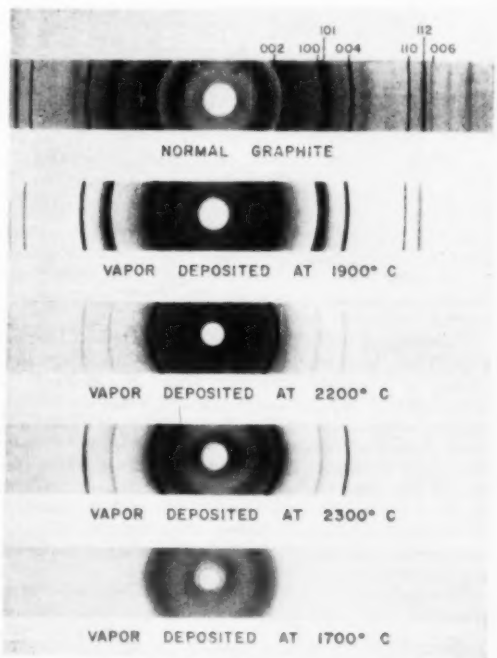

Fig. 1. X-ray photographs of graphite

Table 1. Preparation Data and X-Ray Examination of Graphite

Temperature of deposition (deg. C.)	X-ray data	Method of deposition
2,300	Some disorder in the *c* direction	Resistance furnace 50 mm. mercury
2,200	Some disorder in the *c* direction	Resistance furnace pressure unknown commercial source
1,900	All (*hkl*) reflexions present	Induction furnace 50 mm. mercury
1,700	No (*hkl*) reflexions, lack of three dimensional structure	Resistance furnace 50 mm. mercury
—	Well-defined (*hkl*) reflexions	Commercial graphite substrate material

X-ray diffraction powder patterns obtained differ significantly from results of previously reported investigations. The published literature indicates that as the temperature of deposition decreases, there is a loss of the three-dimensional structure as evidenced by the increasing absence of (*hkl*) reflexions. From Fig. 1, it can be seen that at a deposition temperature of 1,700° C. and pressure of 50 mm. mercury, all the (*hkl*) reflexions are absent. At 1,900° C. there seems to be good atomic alignment in the *c* direction. At higher temperatures of 2,200° C. and 2,300° C., the ordering of the structure again becomes less perfect.

These preliminary observations indicate that high temperature *per se* is not the controlling factor for producing an ordered three-dimensional structure of pyrolytic graphite. The variables of pressure, rate of gas-flow, and method of heating the carbon-bearing gas phase, all have a marked influence on the resultant density and structure. Further investigations are now in progress to determine the effect of these variables on the structure and properties of vapour-deposited material.

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CHEMISTRY

Flash Photolysis of Ferrous Sulphate Solutions

Keen has recently[1] described the kinetics of oxidation of Fe(II) by the transient species produced when a 2-μsec. pulse of 4-MeV. electrons is absorbed by an aqueous solution. We have observed that a similar oxidation occurs in the flash photolysis of ferrous sulphate solutions.

Using a quartz reaction vessel about 1 cm. in diameter and 20 cm. long and a parallel quartz discharge lamp (4 μf., 15 kV.), we find that about 5 μ*M* Fe(III) is produced for each flash irradiation of aerated ferrous sulphate solutions (10^{-4} *M*–5×10^{-3} *M* in 0·01 *N* sulphuric acid) as measured by its absorption at 3130 Å. The effective wave-length in the flash must be less than 2000 Å. since the solutions do not absorb appreciably above this wave-length, and furthermore we find that the oxidation is prevented when an atmosphere of nitrous oxide surrounds the flash tube and reaction vessel.

The oxidation does not result from the direct absorption by Fe(II), since the extent of oxidation is almost independent of concentrations from 10^{-4}–10^{-3} *M*, although it increases at higher, and falls off at lower, concentrations. Moreover, the oxidation

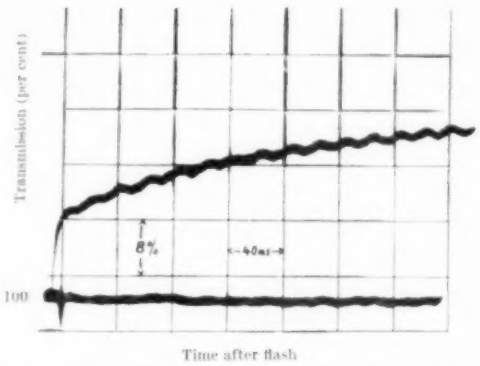

Fig. 1. Optical absorption of 10^{-4} *M* ferrous sulphate in 0·01 *N* sulphuric acid at 313 mμ after illumination with a 40-μsec. flash. Time-scale, 40 msec. per division

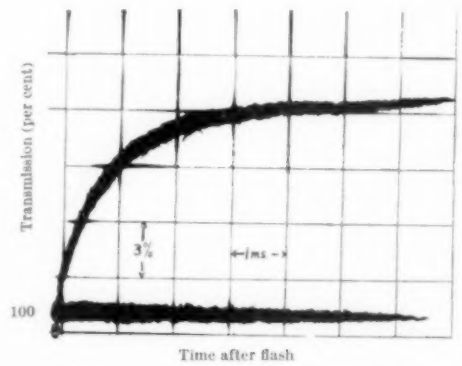

Fig. 2. As Fig. 1. Time-scale, 1 msec. per division

occurs in a time-period well outside that of the flash. We have followed the reaction using the usual photomultiplier-oscilloscope technique and find that at low concentrations there are three distinct stages. Fig. 1, using a time-scale of 40 msec. per division, shows a rapid initial absorption followed by a slower one. With a time-scale of 1 msec. per division the rapid rise is resolved into the curve of Fig. 2. The third stage follows these and extends over seconds at 10^{-4} *M* Fe(II).

A precise kinetic analysis of each of these processes is difficult since they overlap to some extent, but each appears to be first order in the species which oxidizes Fe(II). The slowest step is also first order in Fe(II) concentration (Table 1) and has a second-order rate constant which is exactly that for the $Fe^{2+} + H_2O_2$ reaction[2] (60 ± 2 $m.^{-1}$ $l.sec.^{-1}$). The second stage is kinetically similar to a second-order constant 6×10^4 $m.^{-1}$ $l.sec.^{-1}$, but the first stage is apparently zero order in Fe(II) concentration (Table 1).

The similarity of rate constants suggests that the species oxidizing Fe(II) in the final step is hydrogen peroxide. The other two are more difficult to identify

Table 1. First-Order Rate Constants for the Three Stages of Oxidation of Fe(II) in Aerated 0·01 *N* Sulphuric Acid

Fe(II) m*M*	k_1 $sec.^{-1}$	k_2 $sec.^{-1}$	k_3 $sec.^{-1}$
0·10	370	6·0	—
0·20	380	12·3	—
2·0	—	—	0·120
5·0	—	—	0·310

but presumably one of them, possibly HO_2, is a precursor to the hydrogen peroxide, for example, by the reaction:

$$Fe^{2+} + HO_2 \rightarrow Fe^{3+} + HO_2^-$$

If, as we suggest, absorption by Fe(II) is not the primary reaction, the species oxidizing Fe(II) must arise from absorption by one or more of water, sulphuric acid and oxygen. The effective wavelength cannot be much below 1850 Å. because of absorption by the quartz, and if we ignore oxygen for the moment, the absorption coefficients for water[3] and ions from sulphuric acid[4] indicate that about 60 per cent of 1850 Å. light will be absorbed by water in 0·01 *N* sulphuric acid solution. Water is known to be photolysed[5]:

$$H_2O \xrightarrow{h\nu} H + OH$$

and the sulphate ion may ultimately give the same products:

$$SO_4^{2-} + H_2O \rightarrow SO_4^- + H + OH^- \xrightarrow{H_2O} HSO_4^- + OH + H + OH^-$$

The hydrogen atom with oxygen will rapidly give water, so that we might expect HO and HO_2 to be the oxidizing entities.

However, on this basis the extent of the final slow step would be equal to the sum of the two preceding fast ones, whereas we find it is somewhat less than this. This may indicate that other species, possibly resulting from absorption by oxygen, are responsible for at least some of the oxidation. We would also expect both fast steps to be first order in Fe(II) concentration. The zero order found for the first stage could arise from the relatively slow formation[6] of the absorbing species $Fe^{3+}.SO_4^{2-}$ but we found the presence of 0·01 *N* sodium sulphate to have no effect on this rate.

Subsequent work with perchlorates (to be published, J. H. B.) shows that in the absence of oxygen the final slow step is absent and that the rate constant of the second step is unaffected. This suggests that the second step with a rate constant 6×10^8 m.$^{-1}$ l.sec.$^{-1}$ is:

$$Fe^{2+} + HO \rightarrow Fe^{3+} + OH^-$$

If this is the case it would imply that the reaction:

$$HO + HO \rightarrow H_2O_2$$

must have a rate constant $\geq 10^7$ in order for it not to compete with the Fe^{2+} reaction.

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Use of Flash Photolysis to initiate Detonation in Gaseous Mixtures

Tarsim[1] has reported on the apparent heterogeneity of initiation of reaction in gaseous mixtures of acetylene and oxygen sensitized by nitrogen dioxide, when flash photolysis was used to produce a region of homogeneous chemical reaction. The techniques of using flash photolysis to initiate explosions of hydrocarbon–oxygen–nitrogen dioxide systems are well known[2,3]. This apparent heterogeneity has been explained by the appearance of detonation waves over very small distances. It occurred to me that flash photolysis might be used to initiate detonation waves of certain geometries in a current detonation and shock-wave shaping programme in this laboratory.

The results of one-dimensional experimentation are reported here.

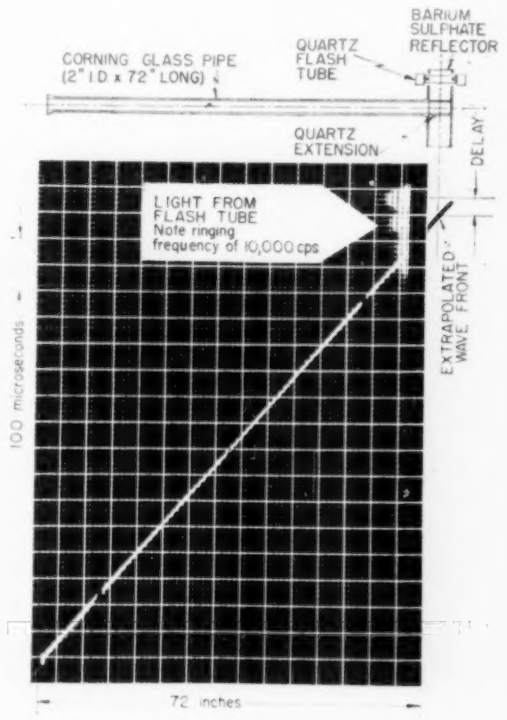

Fig. 1

Fig. 1 gives details of the apparatus employed together with a typical smear photographic record of the detonation wave. A 4-in. length quartz extension was fitted to a 6-ft. length of 2-in. internal diameter Corning glass pipe. The extension was provided with an ultra-violet reflector of barium sulphate and a simple linear quartz discharge tube produced the photolytic flash. The pipe was then oriented with respect to a longitudinal slit so that the detonation wave could be recorded on a simple revolving drum camera. The discharge through the flash lamp in an argon atmosphere was approximately 2,700 joules at 9 kV. The lamp was fired by reducing the pressure of the argon. It will be seen in the smear photograph that the light from the discharge tube was recorded as well as the luminous detonation wave. This gave

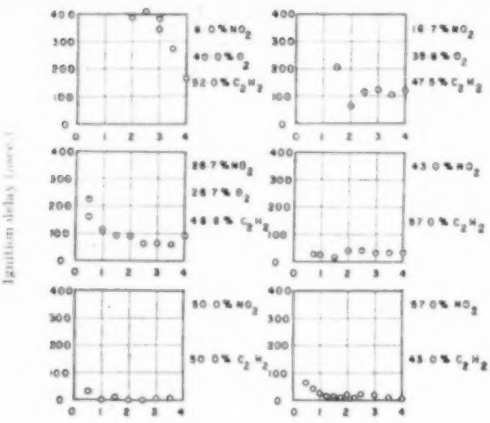

Fig. 2

a measure of the time of the flash with respect to the occurrence of detonation. The time between the first peak in the flash and the occurrence of the extrapolated detonation wave, at the centre of the quartz extension, was arbitrarily taken as the ignition delay.

This ignition delay has been the subject of the early experimentation, and Fig. 2 presents the range of the examination. The variables were the mixture composition and the initial pressure of the mixture. The effect of increasing concentration of nitrogen dioxide was to reduce the ignition delay, while increasing initial pressure had less effect.

In conclusion, stable detonation has been initiated in acetylene, oxygen, nitrogen dioxide mixtures immediately in space, but with a time-delay dependent on mixture composition and initial pressure of the mixture.

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Radiolysis of Carbon Tetrachloride in the Presence of Oxygen

Carbon tetrachloride containing oxygen from the air is very often used as a solvent for the isolation of radioisotopes, as a radiation-polymerization inhibitor, and as a basic material in chemical dosimetry and in sulphur-35 production as well. Its radiolysis is very significant in all these cases.

When carbon tetrachloride in the presence of oxygen is irradiated, free chlorine, phosgene and hexachlorethane are liberated. This system was investigated by Kailan *et al.*[1-3] many years ago, but their results cannot be interpreted quantitatively. Several authors recently investigated this system in the absence of oxygen and the results obtained by them differ greatly from each other[4, 5]. It is impossible to deduce from them the mechanism of radiolysis of carbon tetrachloride in the presence of oxygen. Only Durup[5] occupied himself with the influence of oxygen on radiolysis of carbon tetrachloride, but hitherto nobody has measured the production of phosgene.

We have measured the production of chlorine in carbon tetrachloride irradiated in the presence of oxygen with soft X-rays[10]. Now we present further details about the origin of chlorine and phosgene produced by γ-radiation.

Irradiation was carried out at 22° C. using a cobalt-60 source with a dose-rate of $2 \cdot 5 \times 10^{16}$ eV./min./ml. carbon tetrachloride. Carbon tetrachloride used was a product of Lachema, grade p.a., dry, without further purification. For the measurement of chlorine the spectrophotometer at wave-length 335 mμ was applied. Phosgene was estimated partly by the colorimetric phenylhydrazine modified method, partly by the acidimetric titration. Doses from a cobalt-60 source were measured by the Fricke ferrous sulphate dosimeter ($G_{Fe^{3+}} = 15 \cdot 6$) and a correction on different electron densities was applied.

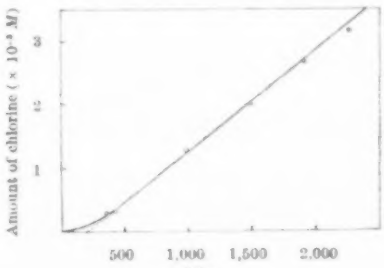

Fig. 1

From Fig. 1 we see that the initial production of chlorine is exponential (induction period up to 140 min. with our conditions) and then linear up to very high doses with $G_{Cl_2} = 3 \cdot 6 \pm 0 \cdot 3$. These results can be explained by the following reactions:

$$CCl_4 \xrightarrow{k_1 I\ CCl_4} CCl_3 + Cl$$

$$Cl + CCl_4 \xrightarrow{k_2} Cl_2 + CCl_3$$

since the calculated concentration of chlorine has the same graphical shape as the experimental curve, then:

$$[Cl_2] = k_1 I t - \frac{k_1 I}{k_2 [CCl_4]} + \frac{k_1 I\, e^{-k_2 [CCl_4] t}}{k_2 [CCl_4]} \qquad (1)$$

Through the analysis of the experimental curve from Fig. 1 by the help of equation (1) we then obtain the rate-constants $k_1 I$ and k_2:

$$k_1 I = 2 \cdot 5 \times 10^{-6} \text{ mole-litre}^{-1} \text{ sec.}^{-1}$$
$$k_2 = 1 \cdot 1 \times 10^{-5} \text{ mole-litre}^{-1} \text{ sec.}^{-1}$$

The G-value of chlorine is higher than in our earlier work[10], because there we used 30-kV. X-rays and the doses were applied in the induction period range.

In Fig. 2 the amount of phosgene is plotted against the radiation dose. We see that the production of phosgene is linear up to the dose 5×10^{19} eV./ml. with the G-value of phosgene $= 12 \cdot 1 \pm 1 \cdot 0$. The situation is rather complicated at high doses and the shape is non-linear. The production of phosgene can be explained by the reactions:

$$CCl_3 + O_2 \xrightarrow{k_3} COCl + Cl_2O$$

$$COCl + CCl_4 \xrightarrow{k_4} COCl_2 + CCl_3$$

$$2\,CCl_3 \xrightarrow{k_5} C_2Cl_6$$

according to which the concentration of phosgene is proportional to the dose:

$$[COCl_2] = \frac{2\,k_1k_3}{k_5}[O_2]\,I^{1/2}t \qquad (2)$$

We are working now on confirming equation (2).

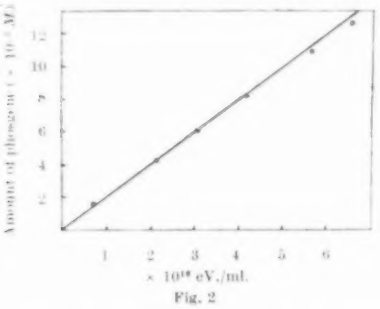

Fig. 2

Both G_{Cl_2} and G_{COCl_2} obtained here are comparable with the results of Durup[9] and other authors[5-7] where $G_{Cl^-} = 3{\cdot}9$ and $G_{-O_2} = 8{\cdot}3 \pm 1{\cdot}0$. According to our reactions the value of G_{Cl^-} must be the same as G_{Cl_2} and approximately $G_{-O_2} \doteq G_{COCl_2}$. For the time being, we have no explanation for the difference existing between G_{Cl_2} in the absence of oxygen[5-8] and in the presence of oxygen.

We are indebted to Prof. Běhounek for his interest and to our co-workers from the Dosimetric Department of the Nuclear Research Institute for their help.

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Liquid Chromatographic Separations using Rubber as the Stationary Phase

In a previous communication[1] it was reported that certain polymers could be separated from mineral oil by a liquid chromatographic method. *n*-Heptane was used as the mobile phase and small pieces of thin vulcanized natural latex as the stationary phase. It has now been found that compounds of lower molecular weight can also be separated by this method.

The use of rubber for the separation of compounds of low molecular weight was first described by Boldingh[2]; he found it advantageous first to swell the rubber with a different solvent from that used as the mobile phase. The use of a cross-linked dextran ('Sephadex') for separating compounds in aqueous solution has been described by Gelotte[3]. Vaughan[4] has mentioned the possibility of using cross-linked polystyrene beads for this purpose.

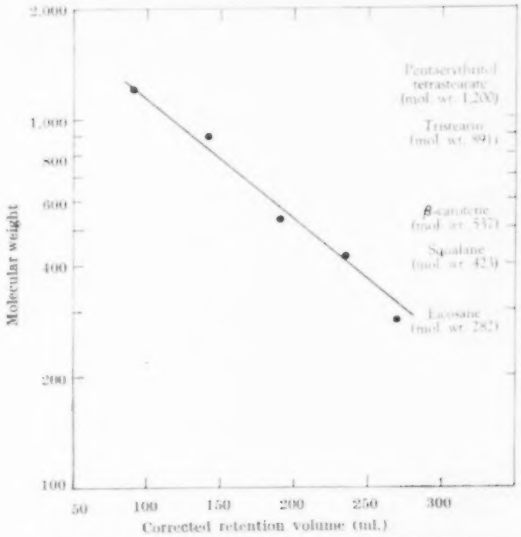

Fig. 1. Effect of molecular weight on corrected retention volume

Boldingh used a fraction of a commercially available rubber powder; but it has been found in these laboratories that columns packed with a similar rubber powder were less efficient than those packed with pieces of surgeons' finger-cots. A column 25 mm.

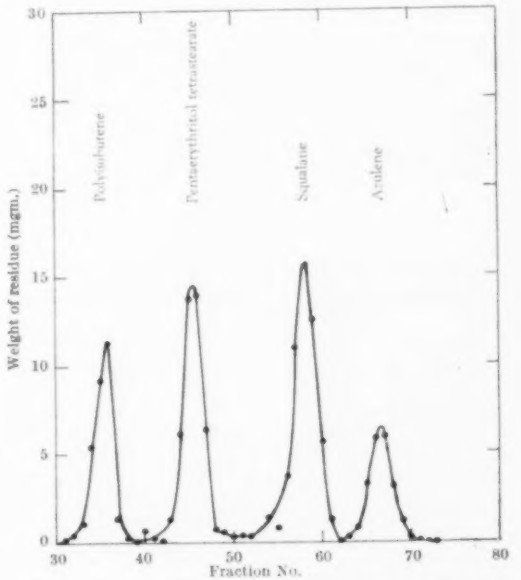

Fig. 2. Separation of a mixture of polyisobutene (mol. wt., 18,000), pentaerythritol tetrastearate (mol. wt., 1,200), squalene (mol. wt., 423) and azulene (mol. wt., 128)

in diameter surrounded by a water jacket at 30° C. was used, packed tightly to a length of 107 cm. with pieces of surgeons' finger-cots passing an 8 mesh B.S. sieve when dry, and retained on a 325 mesh B.S. sieve when swollen with toluene.

The substance (50–100 mgm.) under study was dissolved in 5 ml. of toluene and placed on top of the packing. After the solution had passed into the packing, further toluene was added and the flow-rate adjusted to about 15 ml. an hour. Fractions of 10 ml. were collected in which the solute was determined either by evaporating the toluene and weighing the residue, or colorimetrically in the case of β-carotene and azulene. A graph of corrected retention volume (the retention volume of the elution curve maximum less the void volume of the column) against logarithm molecular weight is shown in Fig. 1. It will be seen that the relationship is approximately linear. An attempt to determine the retention volume of 'Carbowax' polyethylene glycol 1540 was unsuccessful, a very flat elution curve being obtained.

The column used for the above work gave peaks with irregular trailing edges due to the uneven flow of the mobile phase through the column. A second column, 15 mm. in diameter and 207 cm. long, was prepared. Also, the rubber packing was reduced in size so that it passed through a 60 mesh B.S. sieve; and it was packed loosely by allowing it to settle under its own weight. This column gave symmetric elution curves, and the linear relationship between corrected retention volume and logarithm of the molecular weight was again obtained.

The elution curve obtained in the latter column for a mixture of 23·1 mgm. of poly*iso*butene (average molecular weight 18,000), 46·4 mgm. of pentaerythritol tetrastearate, 50·6 mgm. of squalane and 20·8 mgm. of azulene is shown in Fig. 2. Fractions of 5 ml. were collected; recoveries of the compounds ranged from 91 to 103 per cent.

The Directors of Esso Research Ltd. are thanked for permission to publish this communication.

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BIOPHYSICS

Influence of Atmospheric Oxidation on Supercontraction of Wool Fibres reduced with Thioglycollic Acid

The rate of supercontraction of a keratin fibre is progressively enhanced by chemical treatment which results in the breakdown of the disulphide cross-linkages in the fibre. The maximum supercontraction value is, however, not greatly affected by such treatment[1]. Fibres, in which the disulphide cross-linkages have been reduced with thioglycollic acid, contain a high percentage of sulphydryl groups[2]. These sulphydryl groups tend to undergo atmospheric oxidation, which causes the formation of new —S—S— cross-linkages[3], a reaction which can be prevented by blocking the reactive —SH groups. The question arises as to what the effect of such oxidative changes will be on the supercontraction of reduced fibres.

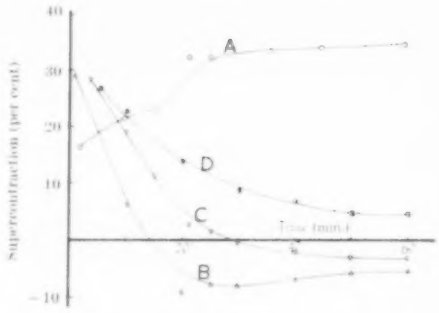

Fig. 1. Fibre reduced for 3 hr. in 1 *M* thioglycollic acid. *A*, normal untreated fibre; *B*, immediately after reduction; *C*, 8 days after reduction; *D*, 20 days after reduction

The microscopic technique, described by Haly and Griffith[4], was employed for obtaining the kinetic supercontraction curves of the fibres.

A sample from a fleece of 64's Merino wool was washed successively in petroleum ether, 0·1 per cent aqueous solution of 'Lissapol N' (Imperial Chemical Industries, Ltd.), water, ethanol and finally ether, all at room temperature, and then dried in a stream of cold air.

Sections of about 1·5 cm. were cut from the root end of a fibre. Snippets, 0·5–1 cm. in length, from both ends of this section, were supercontracted in 7·82 *M* lithium bromide solution at 90° C. If both snippets showed the same supercontraction behaviour, it was assumed that the fibre section was homogeneous along its length. The fibre section was then treated for 3 or 7 hr. at 35° C. with a 1 *M* solution of thioglycollic acid in *p*H 5·2 acetate buffer[5]. After the required reaction-time had expired, the fibre was washed well in oxygen-free distilled water on a sintered glass filter. It was then dried rapidly in a stream of air at 40° C. When dry, a snippet was cut off and supercontracted. The rest of the reduced fibre was stored in the absence of direct light. At various

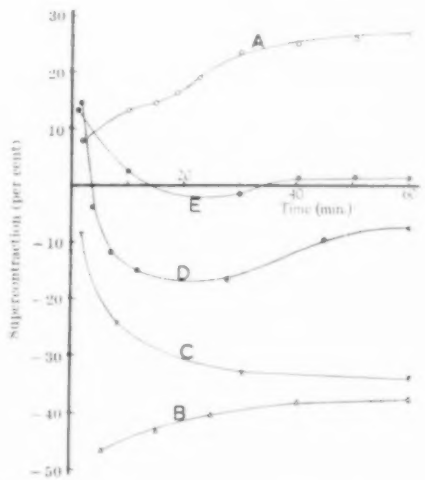

Fig. 2. Fibre reduced for 7 hr. in 1 *M* thioglycollic acid. *A*, normal untreated fibre; *B*, immediately after reduction; *C*, 4 days after reduction; *D*, 10 days after reduction; *E*, 26 days after reduction

time-intervals, snippets were cut off and supercontracted.

The results are shown in Figs. 1 and 2. It is obvious that reduction of 3 hr. is sufficient in the case of Merino wool to break the fibre down to such an extent that partial solution takes place in the contractant. This partial solution could, in fact, be observed under the microscope, particularly at the snippet ends. Due to this partial solution, progressive elongation takes place after the initial rapid contraction. This contraction, which gave values in good agreement with normal equilibrium contraction, was in certain cases so rapid that it could not be photographed effectively at the one-second exposure-time used, and, consequently, no contraction values are shown in the figures. However, as the fibre is progressively exposed to atmospheric oxidation, the extent of partial solution decreases until final contraction values are eventually obtained corresponding to positive contraction. This is probably indicative of the restoration, through atmospheric oxidation, of the disulphide network which is mainly responsible for keratin stability. It would, therefore, be inadvisable to perform supercontraction studies on reduced wool unless precautions are taken to prevent reformation of cross-linkages.

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Ultrasonic Detection of Choledocholithiasis

Present-day methods of detecting gall-stones in the common bile duct at operation are not wholly satisfactory. Stones may be missed in spite of careful palpation, cholangiography and direct exploration of the common bile duct. An attempt has been made to use pulsed ultrasound at the time of operation as an adjunct to their detection.

The apparatus used was a Kelvin and Hughes Mark V *F* flaw detector which is used industrially for the detection of flaws in homogeneous structures such as steel. The detecting probe contains both transmitting and receiving transducers and operates at 2·5 Mc./sec. The transducer emits pulsed ultrasound which is transmitted through a liquid or solid medium with attenuation and is reflected wholly or in part from the surface of any obstruction, the acoustic impedance of which differs sufficiently from that of the medium. The reflexion or echo is picked up by the receiving transducer and the electrical signal generated is amplified and displayed on a cathode ray oscilloscope with a linear time base to give a rough depth calibration (referred to as *A*-scope presentation).

Gall-stones of differing composition (cholesterol, pigment and mixed) and varying in size from 3 to 30 mm. were examined in a tank of saline and definite echoes were obtained from them. They were located by adjusting the position of the transducer head. The optimum range was 3–10 cm. Very slight movement of the transducer produced a marked difference in the size of the echo.

Similar findings were obtained from gall-stones in fresh post-mortem bile ducts and in gall-bladders removed at operation. There were, however, two main differences; the minimum size of gall-stone detectable in bile ducts was 6 mm., and detection proved difficult when the walls of the biliary ducts were grossly thickened by disease. Similar results were obtained when gall-stones in the common bile duct were detected at operation. To provide a liquid medium between the probe and the common bile duct, about 0·5 l. of sterile water was poured into the upper abdomen. The probe was enclosed in a sterile latex sleeve.

Although it is possible with *A*-scope presentation to use ultrasound to detect gall-stones in the common bile duct at operation, the technical difficulties are considerable[1]. It would seem that further research using *B*-scope with plan position indicator[2], which gives a two-dimensional picture, may have something more reliable to offer the surgeon.

We wish to express our gratitude to Prof. W. R. Spurrell, Prof. C. B. Allsopp, Prof. R. Warwick and Prof. G. W. Taylor in whose departments at Guy's and St. Bartholomew's Hospitals the work was carried out. We would also like to acknowledge a grant for this work from the Clinical Research Committee, Guy's Hospital, and to thank Messrs. Kelvin and Hughes for the generous loan of the ultrasonic flaw detector, and the London Rubber Co. for providing the latex sleeves.

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BIOCHEMISTRY

Introduction of the 16α-Hydroxyl Group into Œstrogens by Streptomycetes

Papers concerned with the microbiological transformation of steroids have appeared frequently in the past seven years; however, no examples of the hydroxylation of steroids with a benzenoid *A* ring have been observed. We have been able to introduce a hydroxyl group into the 16α-position of œstrone and œstradiol with two strains of *Streptomyces halstedii* (*ATCC* 13,499 and *NRRL B*-2138) and *Streptomyces mediocidicus ATCC* 13,278.

The cultures were grown for two days in shake flasks at 28° on a medium which contained yeast extract, cerelose, 'NZ amine' type *B* (an enzymatic digest of casein purchased from Sheffield Chemical, Norwich, N.Y.) and calcium carbonate at an initial *p*H of 6·7. This growth in 100 ml. was used to inoculate 2 l. of the same medium contained in a 4 l. fermenter. This mixture was incubated at 28° with stirring at 1,750 r.p.m. and aeration at one volume per volume per minute for 22 hr. After this period, 500 mgm. of œstrone were added in powdered form. The *p*H of the broth was approximately 6·5. The fermentation was continued under the same conditions, for 24 hr. At this time, the whole broth was extracted twice with an equal volume of chloroform. The concentrated extract was qualitatively

examined by means of paper chromatography[1] to determine the transformation products. A ferric chloride colour test was used. Two major transformation products plus œstrone were observed. These products were isolated from the extract by silica-gel partition chromatography in which ethanol was the stationary phase and methylene chloride the mobile phase. The effluent fractions which contained the first more polar compound, were combined. Crystals were obtained, m.p. 214–216°, $[\alpha]_D^{24}$ +174·8 (methanol) λ(KBr, max.) 2·97μ (OH) 5·80 (17-ketone). The melting point and optical rotation are in good agreement with those published for 16α-hydroxyœstrone[2]. The second more polar compound which migrated on papergrams in the œstriol region was also obtained crystalline, m.p. 281–283°, $[\alpha]_D^{24}$ +56 (dioxane). These properties are the same as those reported for œstriol[3]. The infra-red spectrum is identical with that of authentic œstriol.

Estradiol was also converted to 16α-hydroxyœstrone and œstriol. These results indicate the presence of both a hydrogenase and dehydrogenase as well as a 16α-hydroxylase in the above-mentioned micro-organisms.

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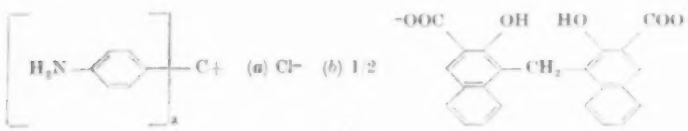
Effects of *Tris*(*p*-Aminophenyl)-Carbonium Salts and Related Compounds on Experimental Schistosomiasis and Paragonimiasis

In 1952, we initiated a programme directed toward the synthesis and biological evaluation of a variety of triphenylmethane derivatives as potential schistosomicides in the hope that a useful curative drug could be developed. To date, more than two hundred have been tested in albino mice and several in rhesus monkeys experimentally infected with a Puerto Rican strain of *Schistosoma mansoni*. Several compounds have also been tested against the lung fluke *Paragonimus kellicotti* in dogs. In the summary presented below, all amounts of drug are expressed as carbonium ion.

In the early stages, we found that a commercial sample of *para*rosaniline hydrochloride (also known as *para* magenta, basic magenta, magenta, parafuchsin (e), *para*-roseine, *CI*-676 (old) and *CI*-42500 (new)) exhibited a lethal effect on both mature and immature worms in mice when fed in the diet and against mature worms in monkeys when administered by gavage. These results demonstrated that a moiety present in the commercial dye might have not only therapeutic but also protective properties.



(I)

Chemical and physical analyses on various commercial samples of the dye indicated that *tris*(*p*-aminophenyl)carbonium chloride (TAC chloride) (I*a*) was usually the major component, although varying amounts of metal salts, *bis*(*p*-aminophenyl)-(4-amino-*m*-tolyl) carbonium chloride, 4,4′,4″-methylidynetrianiline, and N-aryl-*tris*(*p*-aminophenyl)carbonium salts were also present. Since the major moiety was the most likely active principle and such complex mixtures are not suitable for systemic use as drugs, we isolated a highly purified form of TAC chloride. The structure of this compound was substantiated by reduction to 4,4′,4″-methylidynetrianiline which was identical with authentic samples of this material prepared by the condensation of *p*-acetamidobenzaldehyde or 4,4′-diaminobenzhydrol with aniline. Subsequent testing of TAC chloride demonstrated that it had the antischistosome activity previously observed with the commercial dye and, in addition, protective activity in both mice and monkeys.

Although TAC chloride and its base [*tris*(*p*-aminophenyl)methanol](TAC hydroxide) were well tolerated in mice and monkeys at levels necessary for antischistosomal activity, such amounts of both compounds caused emesis and gastritis in dogs. We then searched for other TAC salts[1] which might retain good activity but cause less gastrointestinal reaction and this led to *tris*(*p*-aminophenyl)carbonium salt with 1/2 f.wt. 4,4′-methylene*bis*(3-hydroxy-2-naphthoic acid) (TAC pamoate) (I*b*). It was prepared[1] by allowing purified TAC chloride to react with the disodium salt of 4,4′-methylene*bis*(3-hydroxy-2-naphthoic acid) in aqueous methanol and was isolated as a hydrated green amorphous solid with an indefinite melting point, beginning at 220° C. A representative sample containing 2·5 per cent water of hydration has the following absorption spectrum: ($\lambda_{max.}$ in 50 per cent MeOH–50 per cent *p*H 4·4 phosphate buffer (ε) 237 (133,000), 289 (50,300), 545 (188,000) mμ; $\lambda_{max.}$ in 50 per cent MeOH–50 per cent 0·1 normal sodium hydroxide (ε) 288 (19,400), 365 (6,560) mμ).

Curative studies in mice established that TAC pamoate was active against mature worms over a wide range of well-tolerated doses (200–1,600 mgm./kgm./day) when given orally either in the food for 2–4 weeks or by gavage for 10 days. The dose-response curve was relatively flat. When administered intraperitoneally, the compound was only feebly active. The acute oral LD_{50} in normal mice is greater than 7 gm./kgm. In the monkey, TAC pamoate was well tolerated and suppressed ova production at gavage doses of 100–400 mgm./kgm./day for 10–20 days. TAC pamoate was much less emetic in dogs than TAC chloride.

Inasmuch as 4,4′,4″-methylidynetrianiline is a likely metabolite of the TAC salts[3], it was of interest to compare the schistosomicidal activity of this compound with TAC chloride *in vitro*. TAC chloride proved to be rapidly schistosomicidal *in vitro* at 50 γ/ml. and slowly lethal at 12·5 γ/ml., while 4,4′,4″-methylidynetrianiline was inactive within 72 hr. at these concentrations. These results suggest that TAC salts are schistosomicidal *per se*, but do not, of course, rule out the possibility that there may be active metabolites. Further investigations revealed that the antischistosomal effects of TAC hy-

droxide in mice were not reversed by massive doses of *p*-aminobenzoic acid, whereas the antitrypanosomal activity of commercial pararosaniline in mice is antagonized by the acid[3]. Worms in mice treated with TAC pamoate began to shift to the liver after seven days of medication; death of the worms lagged about six days behind the shift to the liver. Live schistosomes from mice treated for nine to eleven days were stained and examined microscopically for evidence of drug-induced damage. The principal changes were a reduction in the amount of pigment in the cæca of the females and atrophy of the vitellaria. In addition, there was a suggestion of atrophy of the oocytes in the posterior end of the ovaries. The general impression gained from these investigations is that TAC pamoate acts slowly by interfering with nutrition but has little obvious specific organ or tissue toxicity for the schistosomes.

When administered orally to dogs infected with *P. kellicotti*, both TAC chloride and TAC pamoate induced a sustained dimunition in egg production and egg abnormalities, which included shape variations, opaqueness, the appearance of a diffuse germinal mass, and indistinct vitelline cells. When normal adult lung flukes from an untreated dog were incubated for 4 days with 12·5 or 50 γ/ml. of TAC chloride in horse serum, the worms survived but produced similar abnormal eggs; the number was proportional to drug concentration. However, when normal eggs were incubated with 125–500 γ/ml. of TAC pamoate *in vitro*, the only change was a slight staining of the outer covering of the egg. Therefore, it seems likely that these TAC salts act on the reproductive system of these worms rather than on the intact eggs after oviposition.

Among the various triphenylmethanes we have synthesized and tested against *S. mansoni* in mice, the structural requirement for schistosomicidal activity is highly specific. Thus, *tris*(*p*-aminophenyl)-carbonium salts, or derivatives that afford such salts in normal use, such as *tris*(*p*-aminophenyl)methanol and 4,4′,4″-(methoxymethylidyne)trianiline, kill a high proportion of *S. mansoni* in mice. However, curative activity is abolished or drastically reduced when these structures are modified by: (1) reduction to the leuco base (4,4′,4″-methylidynetrianiline); (2) removal of one or two amino-groups; (3) alkylation, halogenation or sulphonation of the benzene rings; (4) alkylation of the amino-groups; (5) removal of one *p*-aminophenyl group; (6) replacement of one or more of the amino groups with a hydroxyl group or a hydrogen or chlorine atom; (7) replacement of the central carbon atom with a sulphur, nitrogen or arsenic atom. These results are in agreement with early reports by Wright *et al.*[4] and Schubert[5] that rosaniline, gentian violet and alphazurine lack appreciable action against mature *S. mansoni*.

During the last stages of this work, Yea-lin and Kuang-sheng[6] reported that rosaniline hydrochloride exhibits certain therapeutic effects on *S. japonicum* infections in mice and rabbits. Recently, Yea-lin[7] revised downward the claims of activity of rosaniline hydrochloride and reported *para*rosaniline hydrochloride to be effective against *S. japonicum* in mice. In several respects our results do not appear to parallel those reported by Yea-lin[7] on the activity of fourteen compounds related to rosaniline against *S. japonicum* in mice. However, uncertainty regarding the composition of the compounds he tested and differences in species and test methods employed preclude a satisfactory assessment of these differences.

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Starch-gel Electrophoresis of Anterior Pituitary Hormones

Separation of pituitary hormones by zone electrophoresis on paper has not proved very successful, possibly due to complex formation between proteins[1]. Starch-gel zone electrophoresis introduced by Smithies[2,3] separates proteins not only by their net charge but also by their molecular size. This procedure has superior resolving power to paper or agar-gel zone electrophoresis and has been successfully applied to the analysis of thyrotrophic hormone[4–6]. Starch-gel electrophoretic analyses of various standard preparations of anterior pituitary hormones are presented here together with some evidence of the distribution of hormonal activities.

A modification of the discontinuous buffer system of Poulik[7] was used. This system was found to separate pituitary proteins better than the original method of Smithies. The electrode vessels contained 0·1 *M* lithium hydroxide and 0·38 *M* boric acid. The gel was prepared with partially hydrolysed potato starch (Connaught Medical Research Laboratories, Toronto) and a buffer containing 90 per cent v/v 0·003 *M* citric acid, 0·016 *M* *tris*(hydroxymethyl)-aminomethane and 10 per cent 0·02 *M* lithium hydroxide, 0·076 *M* boric acid. Electrophoresis was carried out for 4–5 hr. at average voltages of 15–20 V. per cm. During this time sheep albumin moved about 16 cm. and the fastest components in sheep pituitary preparations about 30 cm. from the origin.

The gels were fixed and stained overnight in 0·01 per cent nigrosin (Gurr), in methanol/acetic acid/water 50 : 10 : 40 by volume.

Fig. 1 shows the patterns obtained on the bottom surface of the gels after electrophoresis of various anterior pituitary hormone preparations.

Ox growth hormone (*NIH B2*) has two main components (labelled *A* and *B*) migrating towards the cathode in addition to several minor components migrating to the anode. Several other purified ox and sheep growth hormone preparations have all shown components *A* and *B* but variable minor anodal components.

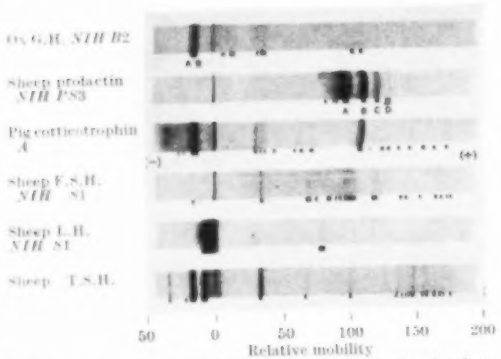

Fig. 1. Photographs of starch gels stained with nigrosin showing the patterns obtained with anterior pituitary hormone preparations. The samples were inserted at the position marked zero on the relative mobility scale and this scale was adjusted so that the prolactin component *A* had a relative mobility of 100

Components *A* and *B* were located in the unstained gel by serrating the edges of the gel with a template as described by Moretti *et al.*[8] and staining a slice from the top of the gel. The pieces of gel containing these components were homogenized with saline and assayed for growth hormone activity in hypophysectomized rats. Both components had approximately equal activity using the width of the tibial epiphysial plate and body-weight as responses. When the components were re-run by inserting the gel sections in a fresh gel they retained their characteristic mobility with no detectable overlapping.

Sheep prolactin (*NIH PS3*), 15 International Units per mgm., shows more than 20 components on the original gel, not all of which can be seen with the amount applied in the experiment illustrated. Purified sheep prolactin preparations from other sources showed similar distribution of the main components. Biological assay in pigeons showed at least 4 regions of the gel, labelled *A*–*D*, having prolactin activity. The main components in regions *A* and *B* in crude sheep pituitary extract have been separated on diethylaminoethyl-cellulose and have been differentially sedimented in intracellular particle preparations from sheep pituitary homogenate (unpublished experiments). However, separation of the components was less readily achieved when purified prolactin preparations were subject to chromatography on diethylaminoethyl-cellulose under the same conditions used for crude extract. Fractions from adjacent peaks after chromatography showed common starch-gel components. This suggests that purification procedures may have increased the tendency of the components to form complexes. A preparation of ox prolactin showed two active components, electrophoretically different from the sheep preparations.

Pig corticotrophin *A*, 108 International Units per mgm., obtained from a pharmaceutical firm, shows a main cathodal component and at least 21 minor components. The cathodal component is common to two other adrenocorticotrophic hormone preparations examined and has a mobility intermediate between the two growth hormone components.

Sheep follicle stimulating hormone (*NIH S1*) shows several minor components, some poorly resolved, but does not have any strongly staining component, suggesting that most of the material in the preparation does not stain heavily with nigrosin.

Sheep luteinizing hormone (*NIH S1*) shows a main cathodal component with very little other material. No biological assay of components in corticotrophin *A*, follicle-stimulating or luteinizing hormone preparations has been carried out.

An impure preparation of sheep thyrotrophic hormone, 1·5 U.S. Pharmacopeia units per mgm., prepared in this laboratory shows at least 15 components. Wynston *et al.*[5] have described four components with thyrotrophic hormone activity in preparations from sheep pituitaries.

The existence of several proteins with one hormonal activity suggests that degradation of the hormone has occurred during fractionation. However, the same components have been found in fresh pituitary extracts subjected to no procedures other than homogenization and centrifugation. Polymorphism does not appear to account for the two active components of growth hormone, since extracts of individual sheep pituitaries all show both components. This hypothesis has not, however, been disproved for prolactin and thyrotrophic hormone. The various preparations examined were obtained from pituitary tissue and it remains to be shown whether multiple forms of anterior pituitary hormones are secreted.

We are grateful to the Endocrinology Study Section, National Institutes of Health, Bethesda, United States, for the supply of growth hormone, prolactin, follicle stimulating hormone and luteinizing hormone. An account of these experiments will be published in more detail elsewhere.

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Polymorphism of α_1-Acid Glycoprotein

α_1-Acid glycoprotein[1,2], a human plasma protein of low molecular weight, has been prepared in a state of extremely high purity as evidenced by electrophoresis and ultracentrifugation over a *p*H range of 1–12 and by terminal amino-acid and immunochemical analyses. Starch-gel electrophoresis of this blood constituent at *p*H 8·5 according to Smithies[3,4] also demonstrated homogeneity. α_1-Acid glycoprotein is further distinguished by the unusually low isoelectric point reported to be at *p*H 2·7 and the high carbohydrate content accounting for 40 per cent and consisting of sialic acid, galactose, mannose, fucose and N-acetyl-glucosamine.

In view of the recently established genetically determined variations of certain plasma proteins[5–8], it appeared of interest to investigate whether or not a comparable polymorphism could be found for α_1-acid glycoprotein. Starch-gel electrophoresis was carried out at the following *p*H values: 7·2, 5·0, 3·8, 2·7 and 2·0. At its isoelectric point this glyco-

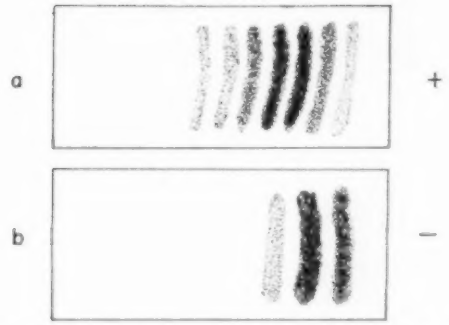

Fig. 1. Starch-gel electrophoresis of α_1-acid glycoprotein at *p*H 2·7 in $\Gamma/2$ 0·02 phosphate buffer (*a*) and of the sialic acid free form of this protein at *p*H 5·0 in $\Gamma/2$ 0·02 acetate buffer (*b*). Position of application of the proteins at the right end of the starch gel. Direction of movement from right to left

protein resolved into several bands, whereas on the acid and alkaline side of *p*H 2·7 homogeneity was observed. A typical preparation[3], derived from 248 litres of pooled outdated normal human plasma (from 1,100 donors of the Commonwealth of Massachusetts), exhibited seven zones (Fig. 1*a*). However, α_1-acid glycoprotein preparations from individual blood specimens separated into different numbers of zones.

To corroborate the validity of these findings, α_1-acid glycoprotein prepared from pooled plasma was treated with neuraminidase, effecting cleavage of essentially the total content of sialic acid, and again subjected to starch-gel electrophoresis. At *p*H 2·7 the resulting protein revealed a single band, but at the isoelectric point of the enzymatically modified glycoprotein, that is, at *p*H 5·0, three zones were noted (Fig. 1*b*). At higher *p*H values apparent homogeneity was restored.

Further work is in progress to determine the genetic implications of these observations.

We are obliged to Dr. J. A. McComb and his associates, Division of Biologic Laboratories, Massachusetts Department of Health, for providing the supernatant solution of Cohn's fraction V derived from pooled normal human plasma, and to Dr. E. H. Eylar, Harvard Medical School, and Dr. E. H. Schultze, Behring-Werke, Marburg-Lahn, Germany, for the gift of neuraminidase.

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Role of Glutathione and of a Self-stabilizing Chain of SH-Enzymes and Substrates in the Metabolic Regulation of Erythrocytes

We have already described the influence of intracellular glutathione on the activity of inorganic pyrophosphatase and triosephosphatedehydrogenase[1,2].

As shown in column 2 of Table 1, other SH-enzymes such as hexokinase, glutathione reductase, glucose-6-phosphatedehydrogenase are inactivated under conditions of oxidation of glutathione by phenylhydrazine according to Beutler[3]. Glucose affords partial protection as may be seen in column 3. The mechanism of the effect of phenylhydrazine is haemin-catalysed oxidation of glutathione in the presence of oxygen[4,5]. Glutathionedisulphide, which is formed as an intermediate, causes oxidation, perhaps by formation of mixed disulphides, and inactivation of the essential SH-groups of the enzymes[6]. The oxidized enzymes, if not reduced, undergo within hours irreversible changes[1,6].

Table 1. Influence of Phenylhydrazine on the Activity of SH-Enzymes

	Control	1	2	3
Incubation (hr.)	0	12	12	12
Phenylhydrazine (5 mgm./ml. suspension)	—	—	+	+
Glucose (2×10^{-2} *M*)	—	—	—	+
Glutathione reduced (GSH) (mgm./100 ml. cells	73·3	64·8	14·6	31·4
Hexokinase ($M \times 10^{-3}$/l. cells/hr.)	18·1	21·9	2·7	23·4
Glutathione reductase ($M \times 10^{-3}$/l. cells/hr.)	26·3	31·0	10·2	15·8
Glucose-6-phosphate dehydrogenase $\Delta E \times 10^3$/0·02 ml. haemolysate/min.	30·2	24·6	7·0	17·8

The determinations of GSH (ref. 16), hexokinase (ref. 17), glutathione-reductase (ref. 18), glucose-6-phosphate dehydrogenase (ref. 19) were performed according to published methods.

Protection of the SH-enzymes is ensured by the reduced triphosphopyridine-nucleotide-forming reactions of the oxidative breakdown of glucose and the glutathione reductase[7]. The interrelation between the glutathione system (GSH/GSSG) and the SH-enzymes are depicted in Fig. 1.

The enzymes, which are concerned with the reduction of glutathione, carry essential SH-groups[8–10]. They are also inactivated when glutathione is oxidized.

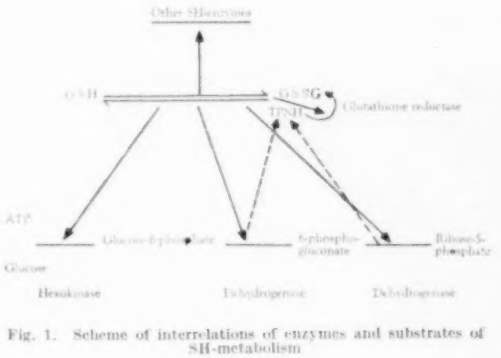

Fig. 1. Scheme of interrelations of enzymes and substrates of SH-metabolism

Thus the extent of enzymatic glutathione reduction is diminished. In case of prevalence of oxidative conditions, more oxidized glutathione would be formed, which in turn would inactivate the SH-enzymes, which participate in the reduction of glutathione. Thus a vicious cycle with destruction of SH-enzymes and glutathione would arise. The same chain of events would occur when one member of the enzymatic self-stabilizing chain of glutathione reduction is missing. Since the chain of SH-enzymes comprises hexokinase, the pacemaker of glycolysis[11], as well as the key enzymes of the oxidative breakdown of the glucose, the oxidation of glutathione might also lead to a decline of the metabolism of glucose and a diminution of formation of adenosine triphosphate with consequent breakdown of cell structure and haemolysis.

It would appear that the system of SH-enzymes and their substrates plays a significant part in the regulation of metabolism and maintenance of structure of the cell, perhaps particularly for the processes of maturation and senescence[12–15].

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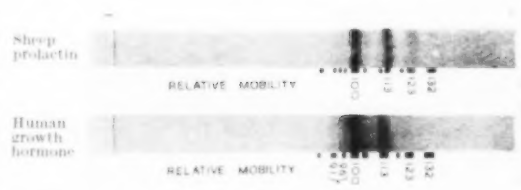
Prolactin Activity of Human Growth Hormone

Zone electrophoresis on starch-gel revealed that purified sheep prolactin contained at least four components with crop-sac stimulating activity in the pigeon[1]. Recently three preparations of purified human growth hormone made by different procedures in different laboratories have been examined by the same method. They all show a striking similarity to sheep prolactin in the mobility and relative amounts of the principal components. This similarity has also been observed by Barrett, Friesen and Astwood[2]. By contrast the patterns are quite unlike the patterns we have observed with ox, sheep, pig and whale growth hormone.



Growth activity percentage recovered 29, 39, 23, 10
Prolactin activity percentage recovered 41, 12, 9, 8

Fig. 1. Starch-gels stained with nigrosin showing patterns obtained with sheep prolactin and human growth hormone

A comparison of one human growth hormone preparation, prepared by Dr. Raben, with the standard sheep prolactin *NIH P-S3* is shown in Fig. 1. Amounts of the two preparations applied to the gel were 2 mgm. for the human growth hormone and 0·5 mgm. for sheep prolactin. Based on staining intensity, these amounts supply approximately the same amount of the principal components in each preparation. Biological assay of the two preparations for crop-sac stimulating activity gave relative potencies of the same order, the human growth hormone being 2·8 International Units per mgm. compared with the prolactin, which is rated at 15 International Units per mgm. The human growth hormone assayed against the ox growth hormone standard *NIH B2*, using the width of the tibial epiphyseal plate in the hypophysectomized rat, was found to be equal to it in potency. If, as may be inferred from the intensity of the components in relation to amount of human growth hormone applied, more purified preparations are possible, then a growth-promoting potency several times that of pure ox or sheep growth hormone may be attainable.

The major components are labelled by their mobility relative to the principal component of sheep prolactin, taken as 100. The amount of prolactin applied to the gel is insufficient to show up the minor components in the photograph. Components 100, 113, 123 and 132 appear in both preparations and correspond to those labelled *A*–*D* which had prolactin activity in the earlier investigation[1]. Components 123 and 132 in sheep prolactin can each be resolved into two components, but this did not occur in the analysis of the human growth hormone preparation.

In another experiment, components 91 and 96 combined, 100, 113 and 123 of the human growth hormone were cut out of the gel, having been located in the unstained gel as described earlier[1]. The amount of gel with each component was equalized by adding blank gel to the narrower components. The separate fractions were assayed for prolactin and growth hormone activity. For both assays single dose levels and six pigeons or rats per fraction were used. The same proportion of the total amount of each component present in the gel was injected. In the same assays were included two dose-levels of standard prolactin, *NIH P-S3* and standard growth hormone, *NIH B2*, respectively, homogenized with the same amounts of starch gel as the separate components. Also included was a single dose-level of the human growth hormone similarly treated.

All the components of human growth hormone tested were found to have both prolactin and growth hormone activity. The only estimate of the relative

amounts of each component at present available is based on staining intensity and may be biased by different affinities of the components for the dye. Accordingly, rather than give doubtful relative potency figures, only the proportion of the activity applied to the gel (corrected for the amount removed in the top section and edges) appearing in each of the components is shown in Fig. 1.

Components 91 and 96 contain a considerable proportion of activity and must have a high potency. The minor components which occur in this region of sheep prolactin have not been tested for prolactin activity. The recovery of activity in the components accounts for somewhat less than the estimated activity applied to the gel for prolactin. However, the discrepancy is within the error of the assay. The ratio of growth hormone to prolactin activity is not constant for the different components, but further bioassays are required to confirm this.

Although the significance of the multiple active components remains to be discovered, it seems likely that the prolactin activity of human growth hormone is not due to contamination, but is a function of the same molecules that possess growth hormone activity.

The similarity of the starch-gel electrophoretic patterns of human growth hormone and sheep prolactin suggests that human growth hormone as now prepared may be chemically and phylogenetically more closely related to the prolactins than to the growth hormones of the ox and sheep, the species most studied in this respect. Further similarity is seen in the chemical and physical properties of the two hormones[3,4]. In molecular weight and amino-acid composition, the growth hormones of different species investigated appear to fall into two groups quite distinct from one another, except that both possess carboxyl and amino-terminal residues of phenylalanine. The group comprising man, monkey and pig is similar to sheep and ox prolactin except in terminal amino-acids, and it is tempting to suggest that monkey and pig growth hormone may also have prolactin activity. However, the starch-gel electrophoretic pattern of pig growth hormone does not resemble that of sheep prolactin.

It is well established that primates do not respond to ox growth hormone[9]. However, it is not certain that a growth hormone related to the hormone of the ox, sheep and whale is absent in primate pituitaries or indeed in the pig. For example, Wilhelmi's method[6,7] for the extraction of ox growth hormone applied to human pituitaries[13] leaves behind much of the growth activity which can then be extracted with solvents which normally extract sheep prolactin. It is possible that some of the growth hormone initially extracted is not identical to that which is brought out by the later extraction.

Sheep prolactin has been shown to have growth hormone-like effects on glucose-tolerance and insulin-sensitivity in dogs[8] and monkeys[9], while stimulation of nitrogen retention in humans[10] and body growth in pigeons[11] and rats[12] has been reported. Such evidence makes it less surprising that human growth hormone preparations should possess both growth hormone and prolactin activity, and suggests that the metabolic effects of prolactin may be a more important part of its function than has been generally recognized.

We are indebted to Dr. M. S. Raben for the gift of human growth hormone and to the Endocrinology Study Section, U.S. National Institutes of Health, for the gift of sheep prolactin.

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Nitrogen and Sulphur at the Active Centre of Carboxypeptidase *A*

A series of metallocarboxypeptidases has been prepared by the substitution of manganese, cobalt, nickel, copper, cadmium, or mercury for zinc, the element present in and essential to the enzymatic activity of native carboxypeptidase *A* [1]. The stability constants, peptidase and esterase activities of these derivatives have been studied and compared[1–3]. Zinc is bound to the single sulphydryl group of the apoenzyme[4–6], as is cobalt as evidenced by its exchange with zinc and by the spectral changes which accompany its binding[2,5]. It has been suggested that a sulphur and a nitrogen atom of carboxypeptidase, together with a metal atom, constitute the active centre of the enzyme, their interaction being required for activity[5].

We now wish to present further evidence in support of the hypothesis that a nitrogenous group is involved in the binding of metals to the active site of carboxypeptidase *A*. The stabilities of the complexes of these metals with a series of nitrogen-sulphur bidentate ligands correlate closely with the affinity of these metals for the binding site on apocarboxypeptidase. The apparent stability constants of the metallocarboxypeptidases in Table 1 (col. 1) have recently

Table 1

Metallocarboxypeptidase	1	2
[(CPD)Me]*	Log stability constant (app.,†)	Log stability constant (corr.)‡
[(CPD)Mn]	5·6	5·6
[(CPD)Co]	5·8	7·0
[(CPD)Ni]	5·7	8·2
[(CPD)Cu]	5·1	10·6
[(CPD)Zn]	8·3	10·5
[(CPD)Cd]	7·5	10·8
[(CPD)Hg]	6·7	21·0

* 1 gm. atom of methyl per mole of protein.
† Determined in 1 *M* sodium chloride, 0·05 *M tris*, *p*H 8·0, 4° C.
‡ Corrected for the reduction of (Metres) by the competing *tris* and Cl^- complexes.

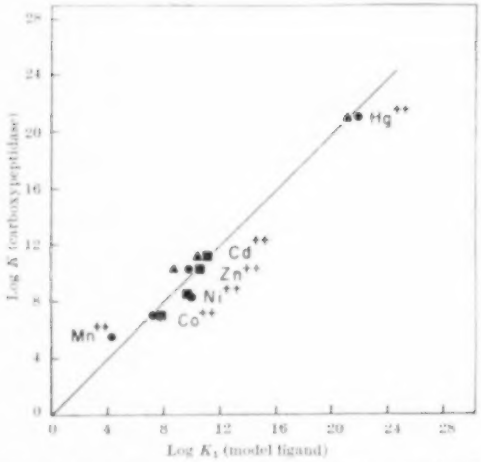

Fig. 1. Correlation of log $K_{corr.}$ for carboxypeptidase with log K_1 for sulphur–nitrogen ligands. ●, cysteine; ■, mercaptoethylamine, corrected for the presence of 0·1 *M* KCl; ▼, glutathione. All log K_1 values are taken from ref. 7

been measured at *p*H 8, 4° in 0·05 *M tris*(hydroxymethyl)aminomethane and 1 *M* sodium chloride, the latter required to dissolve the enzyme, which is otherwise quite insoluble. In these circumstances, zinc is bound more firmly than any of the other metals studied. Allowing for the complexation of the different ions by *tris* and chloride, an appropriate correction must be made for each to ascertain the true stability constants. As is evident from Table 1 (col. 2), this correction is particularly large for mercury. We have compared the stability of these metallocarboxypeptidases with similar data from the literature[7] for cysteine, glutathione, and 2-mercaptoethylamine, which form complex metal ions with nitrogen and sulphur as the donor atoms (Fig. 1).

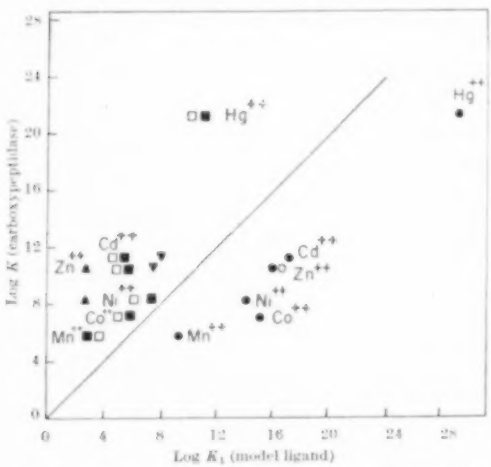

Fig. 2. Lack of correlation of log $K_{corr.}$ for carboxypeptidase with log K_1 for nitrogen–oxygen, nitrogen–nitrogen, sulphur–sulphur, and sulphur–nitrogen–oxygen–oxygen ligands. □, glycine; ■, ethylenediamine; ▲, α-mercaptobutyric acid; ▼, mercaptoacetic acid; ●, 2-mercaptoethyliminodiacetic acid; ○, dimercaptosuccinic acid. All symbols refer to the ion appearing on the same horizontal level as the symbol, except for Cd^{++}, which is slightly displaced from the symbols in both instances. All log K_1 values are taken from ref. 7

The high correlation of the stability constants of the metallocarboxypeptidases with those of the complexes of the corresponding metals with these nitrogen–sulphur ligands is striking, and implies a common denominator. We believe that these results strongly support the hypothesis that, together with the single —SH group of the enzyme, a nitrogen atom is involved in binding zinc and other metals to the apocarboxypeptidase.

In Fig. 2 these enzyme stability constants are plotted against those of nitrogen–oxygen, nitrogen–nitrogen, and sulphur–oxygen bidentate ligands as well as a sulphur–nitrogen–oxygen tetradentate ligand. In contrast with Fig. 1 there is marked scatter of the data ruling out any of the modes of binding shown by these ligands as being consistent with that of carboxypeptidase *A*. The constants for disulphide ligands are orders of magnitude higher than those of carboxypeptidase, as exemplified by the chelate of zinc with dimercaptosuccinic acid, also shown in Fig. 2.

It is therefore suggested that the active centre of native carboxypeptidase *A* includes one sulphur, one nitrogen, and one zinc atom. Present evidence indicates that the nitrogen atom is either part of an α-amino or, perhaps, an imidazole group[3]. Studies to differentiate between these are in progress.

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Similarity of Peptides in Two Antigen-combining Fragments released from a Rabbit Antibody by Papain

During an examination of the enzymatic hydrolysis of rabbit γ-globulin, it was noted that only one-half to two-thirds as many peptides could be differentiated in the hydrolysates as was expected from the amount of base needed to maintain *p*H during hydrolysis: approximately 50 peptides were distinguished following hydrolysis of rabbit antibodies with trypsin, 70 following chymotrypsin and 100 following subtilisin, whereas approximately 100, 150 and 210 μmoles of sodium hydroxide per μmole of antibody respectively were required to maintain *p*H at 8·0. The results suggested the possibility that similar or identical peptides were released from different parts of the same molecule resulting in a smaller number of

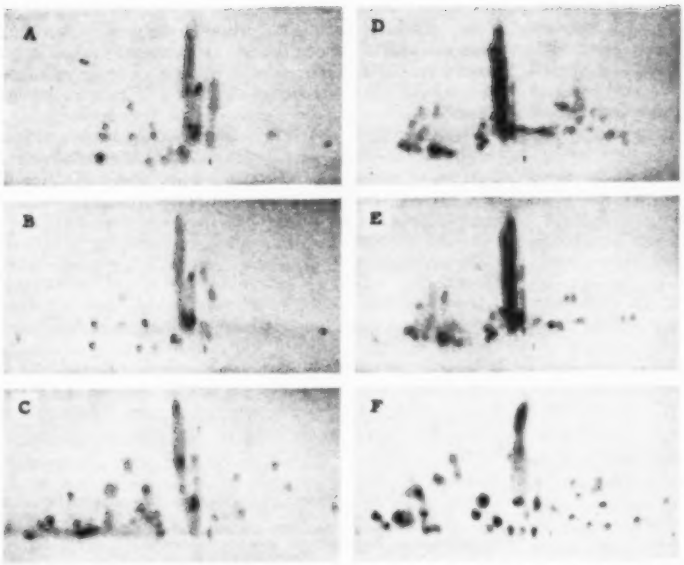

Fig. 1. Peptide patterns of chymotrypsin hydrolysates of I (*A*), II (*B*) and III (*C*) and of the subtilisin hydrolysates of I (*D*), II (*E*) and III (*F*). In each figure, the length of the vertical stroke demarcating the origin at the bottom centre was 7 mm., the anode was to the right and solvent chromatography was in vertical direction

detectably different peptides than the number of peptide bonds broken. An opportunity to test this hypothesis was afforded by Porter's isolation of three large peptide fragments from papain hydrolysates of rabbit γ-globulin[1]. Two of these, I and II, had antibody characteristics and together comprised 50–60 per cent of the antibody molecule; the third fraction, III, was easily crystallized but had no antibody activity. In the work reported here, the peptides of these three fragments were compared.

Rabbit antibodies against type VII *Pneumococcus* polysaccharide were isolated from pooled type-specific rabbit γ-globulin by precipitation with homologous antigen. The specific precipitate was washed four times with 0·15 *M* sodium chloride, suspended in 0·1 *M* potassium chloride containing 0·01 *M* cysteine and 0·002 *M* sodium ethylene-diaminetetraacetate and hydrolysed in the *p*H-stat[2] at *p*H 7·0[3] with twice-crystallized papain in an enzyme to antibody ratio of 1 : 1,000 by weight. The hydrolysate was kept at 2° C. for 2 days and the crystals which formed, fraction III, were collected and recrystallized twice. The remainder of the hydrolysate was dialysed against distilled water and fractionated on a carboxymethyl cellulose column using a *p*H 5·5 acetate gradient, 0·005–0·9 *M*[4]; all column fractions other than I and II were discarded. Type VII polysaccharide was readily detected immunochemically in I and II but not in III. In accord with this, S_{w20} for III in the ultracentrifuge was 3·5 while I and II sedimented much more rapidly, the sedimentation constant depending on the ratio of antibody to antigen in the specific precipitate; the rate of sedimentation of I and II could be decreased by the addition of type VII polysaccharide, in keeping with a decrease in the size of an antigen-antibody complex.

The three fractions were oxidized with performic acid[5] and hydrolysed in the *p*H-stat at *p*H 8·0 with chymotrypsin or subtilisin. 0·1–0·4 mgm. of peptides were applied to a 23 × 57 cm. sheet of Whatman No. 1 filter paper and electrophoresis performed at 52 V./cm. for 45–50 min. at 21–28° C. under 'Varsol' (Esso Standard Oil Co.) at *p*H 4·2 (0·5 per cent pyridine in 0·2 *M* acetic acid). The papers were dried and subjected to ascending chromatography with 2,4,6-collidine, 2,6-lutidine and water in a volume ratio of 1 : 1 : 1. The peptide patterns were developed with ninhydrin.

Approximately 35 peptides could be differentiated in the chymotrypsin hydrolysates of I and II (Fig. 1, *A* and *B*) and about 50–55 in those of III (Fig. 1 *C*). The peptide patterns obtained for the subtilisin hydrolysates of I and II revealed 55 to 60 peptides (Fig. 1, *D* and *E*) and 70 to 75 peptides in the subtilisin hydrolysates of III (Fig. 1 *F*). Per μmole of I, II and III[5], 35, 35 and 60 μmoles of sodium hydroxide were used respectively during chymotrypsin hydrolysis and 60, 60 and 90 μmoles during subtilisin hydrolysis.

As can be seen from Fig. 1, the peptide patterns for I and II hydrolysed under the same conditions were almost identical; a few reproducible differences were apparent. The patterns for III differed quite markedly from those of I and II, but overlapping of some peptide positions in hydrolysates of III with those of I and II was noted.

It would appear, therefore, that approximately half the spots in the patterns obtained for total antibody hydrolysed with chymotrypsin or subtilisin contained peptides derived from at least two different parts of the same molecule. Since rabbit γ-globulin appears to have a single peptide chain[5], it is interesting that two large portions, each equivalent to almost one-third of the chain and each containing an antigen-combining site of the antibody, should have very similar peptide structures.

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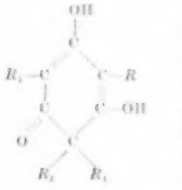
'Sunlight' Flavour of Beer

The fact that an unpleasant flavour occurs on exposing beer to sunlight was first reported by Lintner in 1875[1]. Since that time much work has been carried out on this phenomenon. No explanation, however, has yet been given.

We have observed by quantitative determination that volatile mercaptan was formed in beer exposed

to sunlight[2]. It has been found that humulones (*a*, *b*, *c*) and lupulones (*d*, *e*, *f*) present in hops are related to the formation of 'sunlight' flavour[3]. Furthermore, it has been observed that 'sunlight' flavour was formed even when humulone or lupulone was added, without being heated, to a fermented solution of sucrose or Japanese *sake*[4]. Among the synthetic lupulone analogues (*g*, *h*, *i*, *j*) only acetolupuphenone (*g*) caused sunlight flavour as does humulone and lupulone (ref. 4 and unpublished results).



a, *d*, $R = (CH_3)_2 CH \cdot CH_2 CO-$
b, *e*, $R = (CH_3)_2 CH \cdot CO-$
c, *f*, $R = CH_3 \cdot CH_2 \cdot (CH_3) \cdot CH \cdot CO-$
a, *b*, *c*, $R_1 = (CH_3)_2 C = CH \cdot CH_2 -$
$R_2 = OH$
d, *e*, *f*, $R_1 = R_2 = (CH_3)_2 C = OH$
$CH_2 -$
g, *h*, *i*, *j*, $R = CH_3 CO-$
g, $R_1 - R_2 = (CH_3)_2 C = CH \cdot CH_2-$
h, $R_1 - R_2 = CH_3 \cdot CH_2 \cdot CH_2 \cdot CH_2-$
i, $R_1 - R_2 = CH_3 CH_2 CH_2-$
j, $R_1 - R_2 = CH_3 - CH \cdot CH_2-$

We have now shown that humulone and acetolupuphenone are degraded by photochemical action and that the 3-methyl-2-butenyl group may be split off. 'Sunlight' flavour is recognized when a mixture of humulone or acetolupuphenone and cysteine is exposed to sunlight under conditions in which oxygen in the solution is replaced by carbon dioxide.

3-Methyl-2-butenyl mercaptan (b.p. 125–127° C.) was synthesized by the method of Braun and Plate[5]. This mercaptan produced the typical sunlight flavour of beer when it was added in trace amounts to a fermented solution of sucrose.

On the basis of this experimental evidence, it may be concluded that sunlight flavour of beer is 3-methyl-2-butenyl mercaptan and that it is formed by a photochemical reaction between the 3-methyl-2-butenyl group in the molecules of humulones or lupulones and certain sulphur-containing compounds present in beer.

Further details will be published in *Agricultural and Biological Chemistry, Japan*.

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PHYSIOLOGY

Metabolic Origin of Urinary Methylamine in the Rat

It has been shown[1] that certain monoamine oxidase inhibitors containing the hydrazine moiety (pheniprazine and iproniazid) besides increasing the level of urinary tryptamine[2] and β-phenylethylamine[3] produce a rise of as much as 300 per cent in the amount of urinary methylamine excreted by rats. To demonstrate the origin of this additional methylamine, rats were injected with compounds which, on the basis of their structure, appeared to be possible precursors of this substance. The compounds studied were glycine, sarcosine, dimethylglycine, betaine, creatine and DL-methionine. 100 mgm. of the compound to be tested was made up in isotonic solution to neutral *p*H and injected into the rat intraperitoneally.

To determine the effect of a monoamine oxidase inhibitor on the metabolism of these substances 1 mgm. of pheniprazine was injected intraperitoneally into each of three rats three hours before they received the possible methylamine precursors; a second group injected with saline functioned as controls. The rats (Wistar, male, 250 gm.) were placed after injection in metabolism cages in groups of three, fed on Wayne Lab. 'Blox' and water *ad libitum*. Urine was collected for 24 hr. and the level of both methyl- and dimethylamine in the sample determined as previously described[1]. The levels of methylamine, expressed as percentages of the controls, are shown in Table 1. Levels of dimethylamine are not shown because they were not significantly altered by any of the treatments.

Table 1. Levels of Methylamine in Urine from Rats injected with Possible Precursors of Methylamine in the Presence and Absence of Pheniprazine
(Expressed as per cent of controls (upper and lower values in parentheses))

Treatment	Untreated: methylamine-level	No. of exp.	Pheniprazine treated: methylamine-level	No. of exp.
Saline	100	—	305 (257–375)	4
Glycine	176 (130–250)	6	468 (360–555)	3
Sarcosine	230 (178–280)	4	612 (540–690)	3
Dimethylglycine	108 (100–120)	4	300 (260–356)	4
Betaine	102 (88–124)	3	460 (405–515)	2
Creatine	159 (146–173)	2	1,240 (1,160–1,320)	2
DL-Methionine	101 (88–114)	4	337 (255–405)	5

In the absence of pheniprazine, three compounds, sarcosine, glycine and creatine, produced significant increases in levels of methylamine, sarcosine being considerably more active in this respect than the other two. Betaine, dimethylglycine and methionine had no effect. Injection of pheniprazine raised the level of methylamine about 300 per cent as was shown previously. This increase was not significantly affected by subsequent injections of either dimethylglycine or methionine. It was raised to more than 400 per cent by glycine and betaine, to 612 per cent by sarcosine and to 1,240 per cent by creatine. Urine collected on the second day showed even greater increases with sarcosine (1,800 per cent) and betaine (730 per cent).

According to the present results, sarcosine and creatine (of the compounds studied), appear to be the most likely precursors of methylamine. Sarcosine may give rise to methylamine through the enzyme glycine oxidase first reported by Ratner *et al.*[4]. Deamidination of creatine would produce sarcosine, which may in turn be demethylaminated to yield methylamine and glyoxylate. Should an alternative route of metabolism of sarcosine (namely, sarcosine oxidase) be blocked by pheniprazine the level of sarcosine available for metabolism by way of glycine oxidase would be increased.

Horner and MacKenzie reported that both methionine and betaine may serve as methyl donors for the methylation of glycine to sarcosine[5]. However, in the present work only betaine facilitates production of methylamine in the presence of pheniprazine. This may well indicate that betaine or its biologically

active derivative is the major methylating agent for glycine.

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Ubiquinone and Tocopherol in Birds

Unlike the rat and the rabbit, it seems from nutritional studies made by several groups of workers that the chicken needs little vitamin E, providing its diet contains an adequate supply of selenium and only limited amounts of unsaturated fat[1–3]. Since tissue-levels of ubiquinone in the rat[4] and rabbit[5] are high compared with those given for some other animal species[6] and since these levels have been found to be dependent on vitamin E[4,5], it was of some interest to investigate tocopherol- and ubiquinone-levels in the chicken and some other avian species, with particular reference to those which show considerable differences in their basal metabolic rates. Chickens were reared to 7 weeks on an ordinary commercial ration, and a group of 4 pigeons and 12 canaries were purchased as adult birds from a commercial supplier. Selected tissues from each species were examined for ubiquinone, ubichromenol and α-tocopherol by analytical methods described previously[7].

As shown in Table 1, extremely low levels of tocopherol were confirmed in the young chicken, and the ubiquinone-levels in this species were among the lowest yet observed by us in animal tissues. The tissues of the pigeon and canary contained considerably higher concentrations of both ubiquinone and tocopherol. The level of ubiquinone in the powerful pectoral muscle of the pigeon is particularly interesting in view of the very high metabolic activity of this tissue (which is known to contain high levels of other enzymes and co-factors concerned with oxidation): it is quite an exceptional concentration compared with those found in muscles of other species we have examined. The uniformly high levels of ubiquinone in all the tissues of the canary can also be related to the high basal metabolism in this species[8]. On chromatographic examination, all the tissues of these birds, which were fed ordinary mixed diets, were found to contain only ubiquinone-50. However, other ubiquinones could easily be introduced into the tissues of the birds by feeding them diets containing yeasts, which contain lower isoprenologues of ubiquinone-50: in the latter case there was a marked difference in the isoprenologue pattern of the various avian tissues.

Table 1. Ubiquinone (UQ), Ubichromenol (UC) and Tocopherol (T) in the Tissues of Some Birds

Species	Tissue	UQ (μgm./gm.)	UC (μgm./gm.)	T (μgm./gm.)
Fowl	Heart	18	3	1·4
	Liver	13	1	0·9
	Breast muscle	1·4	0·2	1·5
Pigeon	Heart	192	12	6·8
	Liver	47	not detected	15·4
	Breast muscle	136	8	0·6
	Gizzard	3	0·2	1·4
Canary	Heart	242	17	17·9
	Liver	52	3	8·0
	Breast muscle	38	3	5·0
	Gizzard	43	4	3·9

Although these results are preliminary in some respects, they are not without significance, and tend to support the concept that there is a relationship between tocopherol, ubiquinone and oxidation processes in animals. Although the three species examined were not on identical diets and the latter may have differed somewhat in vitamin E content, this is a factor that, as Edwin *et al.*[4] have shown, has only a minor bearing on the concentration of tocopherol accumulated by the tissues. The differences described in Table 1 undoubtedly reflect real species variations. The role of vitamin E and selenium in the control of ubiquinone in avian species will be discussed in a more detailed report elsewhere.

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Metabolism of *Gerbillus pyramidum*

The genus *Gerbillus* comprises about thirty species which are distributed throughout Africa. *G. pyramidum* or the greater Egyptian gerbil, as it is also called, is found in Arabia, across North Africa to Morocco on the west and down eastern Africa through the Anglo-Egyptian Sudan to Kenya and Tanganyika[1].

The species discussed in this communication, *G. pyramidum*, differs somewhat from the Mongolian gerbil, *Meriones unguiculatus*, in that it is a larger and heavier animal with a less-docile temperament.

Because there are geographical and climatological differences in the habitats of the two species, inherent differences in their energy metabolism and body temperature regulation might also occur.

Oxygen consumption was measured by means of a closed circuit apparatus which incorporated a commercial recording servo-spirometer (Mouse Spirometer, model 160, Custom Engineering and Development Co., St. Louis, Missouri).

Twenty-one animals were used in the metabolic phase of this investigation, ten males and eleven females, ranging in weight from 72 to 145 gm. The conditions under which the oxygen consumption was measured are those described in a previous paper dealing with the Mongolian gerbil, *Meriones unguiculatus*[2]. The animal was housed, during the test

period, in a small glass chamber, which contained soda lime for absorption of carbon dioxide. The chamber was immersed in a thermostatically controlled water-bath which could be set at any desired ambient temperature. After the animal was put in the chamber, an equilibration period of at least 1 hr. was allowed to elapse before the experimental period was begun. The experimental period lasted about 50 min. At least two consecutive runs had to agree closely before the resultant value was accepted. Metabolic measurements were made at ambient temperatures of 15–40° C. and expressed as oxygen consumption in ml./gm. body-weight/hr. This measurement should be regarded as being that of the resting metabolism, since the animals were not in a fasting state.

Least squares fitting of curves to the data, for temperatures below 30° C., yielded the equation $X = 3{\cdot}88 - 0{\cdot}102\ Y$, where Y represents ambient temperature, and X oxygen consumption in ml./gm. body-weight/hr. The correlation coefficient of this curve is 0·904. The critical temperature for *G. pyramidum* seems to be 30° C. Below this temperature, metabolism increases in a linear fashion, with decreasing ambient temperature, at the rate of 0·102 ml. oxygen/g.n./hr./d·g. C.

Oxygen consumption rises sharply at temperatures above 30° C. The regression line between 30 and 40° C. is $X = -3{\cdot}92 + 0{\cdot}146\ Y$. Between 30 and 40° C. there is a somewhat greater scatter of the results for each of the temperatures at which metabolic measurements were made. As a result the correlation coefficient for the regression line is 0·611.

It was not feasible to measure simultaneously the body temperature and oxygen consumption. Therefore, in order to ascertain thermo-regulatory capacity, it was necessary to expose another group of animals to ambient temperatures similar to those encountered in the metabolic chamber. Five animals were used in this phase of the investigation.

The gerbils were individually housed in large open-top plastic boxes in the hot room. Water was provided, although very little, if any, was consumed. Rectal temperatures were taken with a small thermocouple just before entering the room, and thereafter at hourly intervals. The probe was inserted to a point approximately level with the lower lobe of the liver. This point in our experience approximates the true reading of core temperature. The ambient temperatures chosen were 35 and 40° C. Table 1 summarizes the results of these exposures. The 2-hr. value is included both to enable comparison with the other species, *M. unguiculatus*, and to indicate the thermal state of the animal at the end of a metabolic measurement which usually took 2 hr.

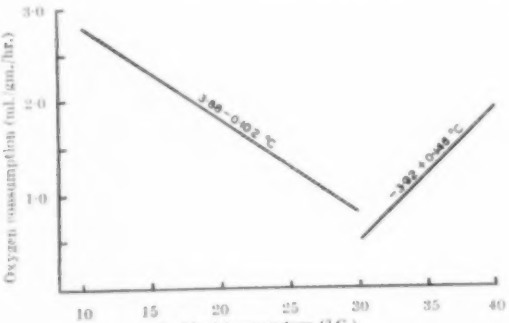

Fig. 1. Effect of ambient temperature on oxygen consumption in *Gerbillus pyramidum*

Table 1. SUMMARY OF THE RESULTS OF EXPOSURE OF GERBILS TO HEAT

Ambient temperature (° C.)	Body temperature (° C.)					
	Mean control at 23° C.	After exposure to heat			Change		
		2 hr.	4 hr.	6 hr.	2 hr.	4 hr.	6 hr.
35	38·7	39·4	38·7	39·1	0·7	0·0	0·4
40	38·1	40·9	42·6	—	2·8	4·5	—

It is clear that while the African gerbil is not greatly inconvenienced at 35° C., even after 6 hr. exposure, it is incapacitated at 40° C. within a very short time. Visual observation alone confirms this. The animals salivate and wet their chests and lie down in a sprawled position, frequently shifting their position as if to seek a cooler location. This behaviour is much like that described by Schmidt-Nielsen[3], Herrington[4] and others[5,6] for the common laboratory animals. It is in sharp contrast to the behaviour of the Mongolian gerbils, which at a similar temperature did not appear to be uncomfortable even though they were exposed for a longer period. Two of the African gerbils exposed to 40° C. died in less than 3½ hr.; two others which seemed severely affected were removed to a cool environment at 4 hr., but died later. Their body temperature at the time they were removed had reached 43·5°.

It is evident from the foregoing results that the metabolic pattern of the African gerbil, *G. pyramidum*, is similar to that of the albino rat. In both animals the resting metabolism is increased between two- and three-fold as the temperature is decreased from 25–10° C. The critical temperature for both species is about 30° C. with a very narrow zone of thermoneutrality. This result is in accord with the pattern for tropical mammals as discussed and illustrated by Scholander[7]. The present results are unlike the metabolic pattern displayed by the Mongolian gerbil, *M. unguiculatus*. The Mongolian species shows an increase in metabolism between 25 and 10° C. (similar to that of the African species) but quite unlike the African species *M. unguiculatus* has a zone of thermal neutrality extending from 30–40° C. This difference between the two species may be due, in great part, to the superior capacity of *M. unguiculatus* to regulate its body temperature in a hot environment. It is likely that *G. pyramidum* spends the daylight hours in burrows where the conditions of temperature and humidity do not tax the animal's heat-regulatory mechanism.

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Spectral Sensitivity of Single Visual Cells

SINCE the work of Young and Helmholtz, the highly developed colour vision in man and many animals has been thought to depend on several, usually three, retinal receptors with different spectral sensitivity[1,2]. Such a theory is best suited for the explanation of human sensations, of animal reactions, and of electrical responses of the whole eye. It is, however, very difficult to determine the spectral sensitivity of the postulated single receptors using these methods. The existence of different photosensitive pigments has been demonstrated by chemical methods in vertebrates[3], and *in vivo* photometric measurements have shown them for the mammalian retina[4]. But this again does not give us an entirely satisfactory explanation of the principal unit-level functions, and the very successful chemical analyses of the visual pigments do not help us understand colour vision[5].

Hitherto, electrical recordings from single elements in the vertebrate retina have been carried out only on the level of ganglion cells and not on the level of single receptors[5,6]. It is clear from these experiments that in the layer of bipolar cells and in the inner plexiform layer we are dealing with the integrated activity and not with the primary reactions of visual cells. Electrical recording from single visual cells of the blowfly (*Calliphora erythrocephala*) was made successfully, using microelectrodes with a tip diameter of approximately 0·1μ (ref. 7).

On the basis of results of electrophysiological and behavioural experiments, colour vision appears to exist in *Calliphora*, and the spectral sensitivity of the whole eye is well known[8]. The study of the spectral sensitivity of single visual cells in this animal may provide information necessary for the understanding of colour vision in arthropods and might probably provide a model for colour vision in general. Basic properties of the receptor potentials, the mechanism of analysing the plane of polarized light in the ommatidia, and details of the methods are described elsewhere[7,9].

In the present experiments, stimulating lights were monochromatic flashes of equal quanta and of 0·1 or 0·02 sec. duration with an interval of 10 sec. The spectral range 315–681 mμ was covered by a series of narrow-band interference filters with a mean wave-length interval of 19 mμ between filter peaks.

The spectral sensitivity of 39 single visual cells was determined for the wild form of *Calliphora*; additional experiments were performed with the white-apricot mutant, in which the red screening pigment is absent.

All cells in the dorsal part of the eye have a uniform sensitivity curve with one maximum at 489 mμ, $\sigma = \pm 7$ mμ, and a second maximum in the ultra-violet at 345 mμ, $\sigma = \pm 3$ mμ. Three different receptor types were found in the ventral part of the eye: (1) a green receptor similar to that in the dorsal part with one maximum at 491 mμ, $\sigma = \pm 6$ mμ, and a second maximum in the ultra-violet, again at 345 mμ, $\sigma = \pm 1$ mμ; (2) a blue receptor with one maximum in the visible part of the spectrum at 468 mμ, $\sigma = \pm 6$ mμ, and a second maximum again at 345 mμ, $\sigma = \pm 2$ mμ in the ultra-violet; (3) a yellow-green receptor with one maximum at 524 mμ, $\sigma = \pm 7$ mμ, and another maximum also at 345 mμ.

Statistical treatment for the 491-mμ and 468-mμ peaks gave $P_{20} < 0{\cdot}0002$; for the 491-mμ and for the 524-mμ peaks, $P_{19} < 0{\cdot}0002$; for the 468-mμ and 524-mμ peaks, $P_5 = 0{\cdot}0002$. Comparison of the maximum for the dorsal part at 489 mμ with the maximum for the ventral part at 491 mμ gives $P = 0{\cdot}8$. All maxima in the ultra-violet are very close to each other, with P between 0·48 and 0·66.

If one compares sensitivity curves of receptors belonging to the most common type with the 491-mμ maximum by setting the maximum at 491 mμ as 100 per cent amplitude, the curves follow one another with one exception; some of them have a hump or even a third peak at 616 mμ. If the elements are compared according to the absolute value of potential amplitudes, the following rule becomes clear: the lower the response of an element, the more significant becomes the hump at 616 mμ, and only elements with very feeble action potentials show a peak at 616 mμ. This observation shows that the red screening pigment does not absorb light of this wave-length, and in weakly illuminated elements the portion of scattered red light becomes much greater than the portion of light coming through the dioptric apparatus of the ommatidium. On the other hand, except in the case of weakly illuminated elements and in the far red, the screening pigments are not relevant to the shape of the sensitivity curves. It appears from these experiments that *Calliphora erythrocephala* has at least three visual receptors with different spectral sensitivities. All sensitivity curves have a uniform peak at 345 mμ. This corresponds very well to the rather stable position of the β-absorption in all derivatives of vertebrate visual pigments. The correct value of this wave-length may be the same in *Calliphora* and in vertebrates, because our measurements may have been affected by the quality of filters available. It may be assumed that the visual pigment of the most common type of visual cell in *Calliphora* is a retinene with the maximum absorption near 490 mμ. This pigment is already known to occur in insects[10].

All the cells in the upper part of the eye yielded only one uniform curve. This situation is similar to that in other insects, which have been found to have one part of the eye sensitive to colours and another part colour-blind; for example, *Periplaneta*[11], *Notonecta*[12], and *Libellula*[13]. The most common receptor in the ventral part functions like the dorsal one. The numerical relationship among green, blue and yellow-green receptors is 18 : 4 : 3. If the assumption is made that five of the seven visual cells within each ommatidium are green receptors, and, of the remaining two, one is a blue and the other a yellow-green receptor, the numerical relationship would come very close to that actually found.

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A Potential Source of Error in the Hexobarbitone Sleeping-Time Test in Mice

RECENTLY, Bhide[1] directed attention to the effect of hydration on the hexobarbitone sleeping time of mice. We have been able to confirm this work and that of earlier workers with respect to the effects of hydration in mice[2], but it is not generally appreciated that dehydration, whether achieved by means of diuretic agents or deprivation of water for relatively short periods, has a significant effect on hexobarbitone sleeping time; these effects may lead to error in the screening of potential central nervous system depressants where these agents also have some diuretic or antidiuretic activity and where water supplies are accidentally removed from the animals for short periods.

We have found that a series of diuretic substances, mainly unrelated either chemically or in their mode of action, reduced hexobarbitone sleeping time in mice as did deprivation of water for relatively short periods.

Unfasted female albino mice weighing 17–20 gm. received hexobarbitone sodium 100 mgm./kgm. at different intervals after receiving a dose of diuretic. The interval at which maximum effect was obtained is the only one reported. Sleeping time was taken as the interval between the loss and the regaining of the righting reflex. All drugs were given by intraperitoneal route in a volume of 0·2 ml. distilled water per 20 gm. body-weight. The diuretics examined were acetazolamide ('Diamox sodium' 25 mgm./kgm. and 500 mgm./kgm.), sodium sulphate (100 mgm./kgm.), sodium chloride (200 mgm./kgm.), sodium bicarbonate (200 mgm./kgm.), ammonium chloride (10 mgm./kgm.) and injection of 'Mersalyl' B.P. (0·1 ml./kgm.). Control groups received 0·2 ml. of distilled water at an appropriate time-interval before the anæsthetic. Ten mice per group were used throughout.

The results in Table 1 show that all the diuretics used in these experiments significantly reduced the hexobarbitone sleeping time.

Before the administration of hexobarbitone in the water-deprivation experiments, the animals were deprived of water for varying intervals of time but they were not deprived of food. Table 2 shows the results of these experiments. Deprivation of water for 2½ hr. before the administration of the hexobarbitone has a highly significant effect on the sleeping time. Mice have a high turn-over rate of water, about five times that of man[3], and it is reasonable to conclude that they are very susceptible to dehydration whether occasioned by withdrawal of water supplies or by diuretics. A water load of 1·5 ml./kgm., when given intraperitoneally ½ hr. before the anæsthetic, significantly increased the sleeping time.

Table 1

Diuretic	Hours between administration of diuretic and hexobarbitone	Control sleeping time	Sleeping time after diuretic ± S.E.	Values of P (t test)
Acetazolamide, 25 mgm./kgm.	2·0	31·2 ± 3·2	13·4 ± 2·4	0·001
Acetazolamide, 500 mgm./kgm.	2·0	31·2 ± 3·2	10·3 ± 1·7	0·001
Ammonium chloride	3·0	30·1 ± 3·1	20·2 ± 1·8	0·05
Sodium chloride	2·0	26·7 ± 1·6	20·1 ± 1·1	0·01
Injection of 'Mersalyl'	5·0	26·7 ± 1·6	13·5 ± 1·1	0·001
Sodium bicarbonate	0·5	26·7 ± 1·6	18·3 ± 1·2	0·001
Sodium sulphate	2·0	26·7 ± 1·6	15·1 ± 1·7	0·001

Table 2

Hours between deprivation of water and administration of hexobarbitone	Control sleeping time	Sleeping time after water deprivation ± S.E.	Value of P (t test)
0·5	31·5 ± 2·1	27·9 ± 1·2	Not significant
2·5	31·5 ± 2·1	16·9 ± 1·8	0·001

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HÆMATOLOGY

Stathmokinetic (Mitosis-inhibiting) Method for estimating Plasma Erythropoietic Hormone

ASSAY methods for estimating the erythropoietic activity of plasma samples usually involve the evaluation of altered haematological values in test animals, or the change in incorporation of radioiron into the red cells of test animals—usually rats with polycythæmia induced by transfusion. *In vitro* bone marrow techniques do not seem to be widely used.

This communication describes a method for assaying plasma extracts, the stimulating properties of which appear to be confined to erythroid cellular division as observed in the bone marrow of the intact rat.

Activated plasma was obtained by bleeding rabbits (2·5–3·5 kgm.), 10 ml./kgm. body-weight, for 3 consecutive days, to a haemoglobin-level between 5 and 6 gm./100 ml. On the fourth day the animals were exsanguinated under nembutal anæsthesia and blood taken by aortic catheter was received into bottles containing heparin as anticoagulant. The cells were then centrifuged off at —3° C., and the plasma stored at 3° C. Samples were pooled and then extracted, using Lowy and Keighly's modification[1] of the method described by Borsook[2].

Adult Wistar rats, weighing 150–180 gm., from a closed colony were the recipient animals. No animal was used unless it had been in the laboratory for at least 4 days and more usually the period was more than a week.

Injections of plasma extract were made subcutaneously for 3 consecutive days. At 10 a.m. on the fourth day, 18 hr. after the last injection, a subcutaneous injection of colchicine, 0·05 mgm./100 gm. weight, was given. 5 hr. later the animals were killed, a femur removed, and marrow smears prepared. Duplicate smears prepared from each animal were air-dried, fixed in methyl alcohol and stained with May-Grunwald Giemsa stain.

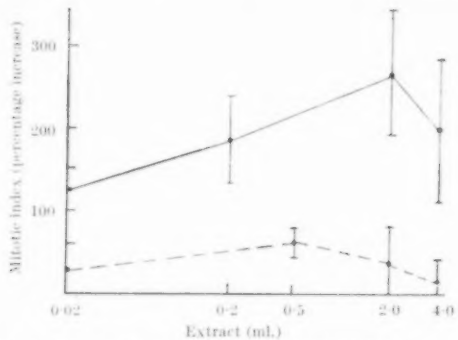

Fig. 1. Response of five groups of rats (4 or 5 rats in each group) injected with extract prepared from the plasma taken from bled rabbits. The interrupted line shows the response to extract from normal rabbit plasma

The films were examined under a high-power lens using oil immersion. Five hundred nucleated red cell precursors were counted, the number of mitotic divisions noted and the result expressed as a percentage (mitotic index). Red-cell precursors at all stages of development were counted, no attempt being made to differentiate between various stages of maturation. Cell differentiation was improved by the florid metaphase patterns resulting from colchicine blocking. A number of cells defied identification. The mitotic index of normal unstimulated animals receiving colchicine was estimated in 8 rats and the mean found to be $3{\cdot}8 \pm 1{\cdot}4$ *S.E.* All results were expressed as a percentage increase above this figure.

Fig. 1 shows a dose response curve using the method in groups of 4 or 5 rats. The interrupted line shows the response with extract prepared from plasma obtained from unbled rabbits. It is at once apparent that a very considerable increase in the mitotic index occurs with very small doses of the active extract. The fall in the response to the 4-ml. (active) dose is what one would expect in such a biological system. The rather sharp fall at 2- and 4-ml. doses for the extract from unbled animals suggests a tachyphylaxis which might result from antibody formation due to injection of relatively large quantities of protein-containing extract of low potency.

Grant[3] used colchicine to demonstrate its mitotic-inhibiting properties in bled rats, and in rats subjected to an atmosphere with low oxygen tension; my results are in accord with his findings.

The main advantage of the method appears to be its sensitivity in the lower dose-range—an advantage in striking contrast to other assay procedures. A major disadvantage is its lack of objectivity—a disadvantage partially removed by re-labelling the specimens by an independent observer, who disclosed the key only when the results were known.

Using this method, I have shown (unpublished work) that although mitotic erythroid hyperactivity is induced, the reticulocyte counts are not altered to a significant degree. Injections continued for 10 days did not produce changes in the hæmoglobin-level; this may mean that the factor necessary to cause this change was lacking, or alternatively that the potency of the material was low and that larger doses than those used (0·75 ml./100 gm.) might be effective. So far as these experiments have proceeded, the only effect has been on the rate of mitosis in the erythroid marrow cells.

It seems probable that this factor responsible for cellular division is identical with that demonstrated by others, for example, the other-soluble fraction described by Linman, Bethell and their co-workers[4]. Further work is proceeding to characterize this factor.

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Paracrystalline Hæmoglobin as a Species Difference between *Rattus norvegicus* and *Rattus rattus*

THE common laboratory white rat is almost always the albino variety of the Norway rat (brown rat, *Rattus norvegicus*)[1,2] and only exceptionally the albino variety of the black rat (*Rattus rattus*)[2]. The two species are morphologically similar, which makes it rather difficult to distinguish them. Nevertheless, there are qualitative characters which make distinction sure.

On suspending the erythrocytes of albino rat in isotonic sodium citrate in a refrigerator their hæmoglobin attains a certain paracrystalline state characterized by general birefringence of these red cells in polarized light (Fig. 1) combined with enormously increased osmotic resistance[1].

Comparative studies showed that the red cells of the brown rat (*Rattus norvegicus*) only, be it the albino or the wild variety, sustain this change. It could not be observed, even after prolonged suspension, in the red cells of either the nearest zoological relative—the wild black rat (*Rattus rattus*)—or other animal species—white mouse, mouse (*Apodemus agrarius*), rabbit, hamster (*Cricetus cricetus*, *Mesocricetus auratus*), guinea pig, cat, dog, horse, pig, horned cattle, sheep and man.

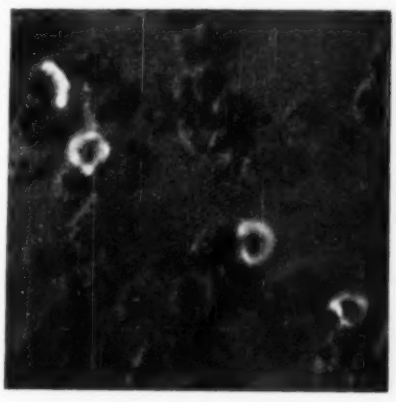

Fig. 1. Diffuse birefringence of paracrystalline red cells in the albino rat (*Rattus norvegicus*) in polarized light after 28 hr. storage in cold isotonic sodium citrate

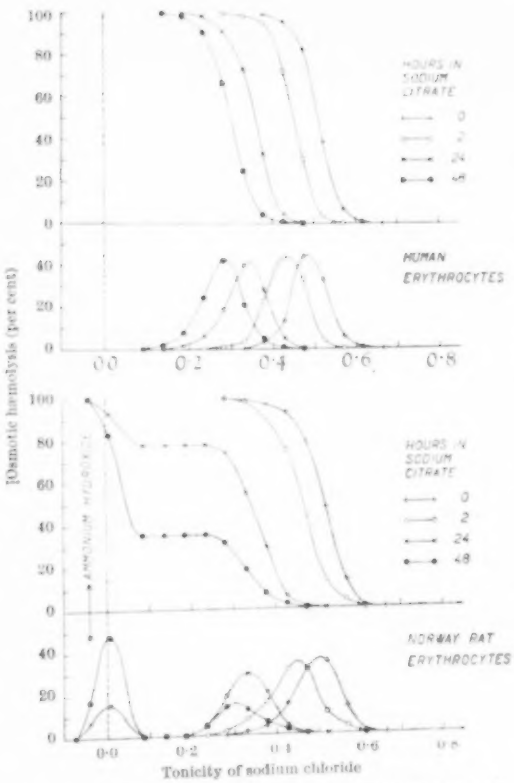

Fig. 2. Integral and distribution curves of osmotic resistance of human and albino rat (*Rattus norvegicus*) red cells during storage in cold isotonic sodium citrate

Similar differences between the erythrocytes of the Norway rat and other animal species were found in changes in osmotic resistance. The storage of erythrocytes in cold citrate is accompanied with gradual and marked increase of their osmotic resistance, especially in the first hours of suspension, and the maximum is attained within 60–72 hr. The increase is even in all red cells as evident from the Gauss-like type of the distribution curves[5]. All but the red cells of the Norway rat react thus, and even extremely resistant cells haemolyse in distilled water (Fig. 2, upper part).

In contrast to that, the osmotic resistance of the red cells in the Norway rat changes in a different way, the cells being unevenly affected by cold citrate (Fig. 2, lower part): one part increases in resistance in roughly the same way as in other species, but the other attains resistance of such a degree that it withstands haemolysis even in distilled water. The respective distribution curves are bimodal, reflecting a change of the originally homogeneous suspension into one with bimodal variability[6].

Two qualitatively different types of red cells thus exist side by side in the citrate suspension of brown rat red cells, the fraction of the paracrystalline birefringent and highly resistant erythrocytes being proportional to the duration of storage in the cold citrate.

The increase in resistance is due to change of colloid chemical properties of haemoglobin proteins resulting from the escape of intracellular colloid water. The relation between mean corpuscular volume of the suspended red cells and the amount of their intracellular potassium and changes in concentration is an indirect proof of this.

The mean red cell volume decreases abruptly during the first 2 hr. of suspension in citrate through the escape of water while the absolute potassium quantity (μμgm. in one red cell) falls much slower. As a result, its osmotically effective concentration (expressed in m.equiv./l. of red cells) is higher than before storage. Red cells become more hypertonic than their environment and the initial decrease of their volume cannot be ascribed to loss of free water which only solves crystalloids but rather to water bound on colloids of the protein red cell core. Through this dehydration of intracellular proteins the red cells become more rigid and gel-like, only swell with difficulty and therefore show increased osmotic resistance.

Red cells with sodium for their main cation (sheep, cattle, cat)[7] and hence having still less possibility of decreasing their osmotically effective intracellular concentration in an environment rich in sodium react in the same way.

Dehydration of intracellular protein is so far advanced in some red cells of the Norway rat that the haemoglobin molecules are forced, owing to lack of room, to arrange themselves into a paracrystalline birefringent space system. The structural orderliness and rigidity of these cells are so great that the cells withstand haemolysis even in distilled water, being unable to swell sufficiently.

The birefringence described and variability of bimodal osmotic resistance in the Norway rat were found in both common laboratory white rat strains and inbred Wistar, *LE*, *PVG* and *WAG* strains. This supports the suggestion that all commonly kept laboratory rats belong to the Norway rat species, not to the black rat (*Rattus rattus*) species.

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PATHOLOGY

Zymograms in Human Pathology

Enzyme histochemistry is sometimes a valuable aid in differentiating tumours in human pathology[1] and in studying several other pathological processes[2].

The methods used in histochemistry are based on the localization of enzyme activity. The action of the enzyme is demonstrated, not the enzyme as such. Moreover, in principle, it is possible to demonstrate any enzyme as a 'substance'. The method of Hunter and Market[3,4], in which tissue homogenates are

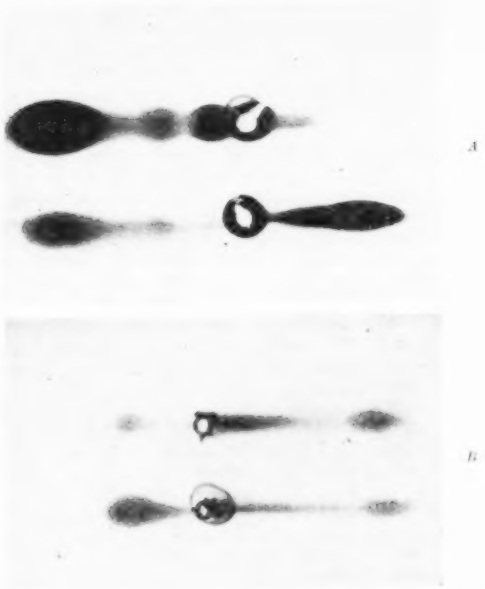

Fig. 1. *A*, Zymogram of homogenate of nodular goitre (upper) and of an adenocarcinoma of the large intestine (lower); *B*, amidoschwarz stained electrophoretic diagrams of the same homogenates ('albumin' moving to the right)

differentiated by means of starch-gel electrophoresis after which the enzymes are stained in the electrophoresis diagram according to a technique derived from histochemistry, offers a possibility of combination of characterizations. The histochemical technique employed locates the action of the enzyme; the position on the electrophoresis diagram more or less characterizes the enzyme as a protein. With the aid of this technique, Market and Hunter[4] succeeded in demonstrating various esterases in homogenates of organs of the mouse. The pictures obtained, indicating the localization of enzymes in the electrophoresis diagram, are called zymograms.

We used a somewhat modified and rather simpler technique, namely, agar electrophoresis[5], followed by histochemical location of the enzyme esterase in various anomalies in human pathology.

Histochemical slides (2·5 × 7·5 cm.) are immersed in 1 per cent agar solution and dried. Next 3 ml. of 1·5 per cent agar solution in veronal buffer *p*H 8·6 is poured on the slides. Beforehand, strips of electrophoresis paper are attached to the ends of the slides by means of buffer. These are hung in the electrode trays. With an injection needle, straight holes are punched in the agar which are filled with homogenate. This is prepared by homogenizing tissue in a Bühler apparatus with equal volume of distilled water. After electrophoresis (4 hr., 5 V./cm.) the electrophoresis slides are stained for 0·5 hr. by Gomori's esterase method[6]. The substrate is prepared by adding 1 ml. of 1 per cent α-naphthol acetate in 50 per cent acetone and 5 ml. of 0·2 *M* disodium hydrogen phosphate to 50 ml. of distilled water. In it some fast blue salt *RR* or *B* is dissolved. After incubation it is washed in distilled water, followed by overnight fixation in ethanol 50 per cent. After drying at 37° C. (glasses covered with filter paper) it is covered with cover-glass and glycerin gelatin. Other glasses are stained for protein after electrophoresis by means of amidoschwarz.

Fresh human material, mostly obtained by operation, was prepared in this manner. Several anomalies in various organs could be studied. Also in human material the zymograms thus obtained show distinctly different esterases. Fig. 1 gives an illustration. In Fig. 2 a few zymograms of anomalies in the digestive tract, thyroid gland and pulmonary tumours are grouped schematically. These illustrations are representative of what was observed in larger material. With our technique, in the main, 6 esterases are encountered in human material (I, II, III, IV, V, VI). Three of these (I, II, III) move in the direction of the albumin from the blood, the other three in the direction of the globulins.

Some general conclusions may already be drawn from our observations. It appears that different organs do not show the same zymogram. There are qualitative differences, for example, in the homogenates of the thyroid gland, the I and II esterases are lacking, and quantitative differences, for example, the predominance of I esterase in the digestive tract. Histochemically the highest esterase activity in thyroid gland, digestive tract and lung carcinoma was localized in the epithelium. The zymograms show highest activity of VI, I and II esterase respectively. The question arises whether electrophoretically different esterases show characteristic sites of localization. Further, it is noteworthy how the tumours of a single organ (lung carcinoma) show a fairly identical pattern. An adenocarcinoma of the appendix is clearly different from it. This displays a pattern similar to some non-tumour-like anomalies of the digestive tract.

Protein staining of the electrophoresis slides naturally shows a pattern quite different from the zymogram. It is noteworthy that protein is often scarcely demonstrable at the site of highest enzymatic activity (Fig. 1). This opens up possibilities for isolation.

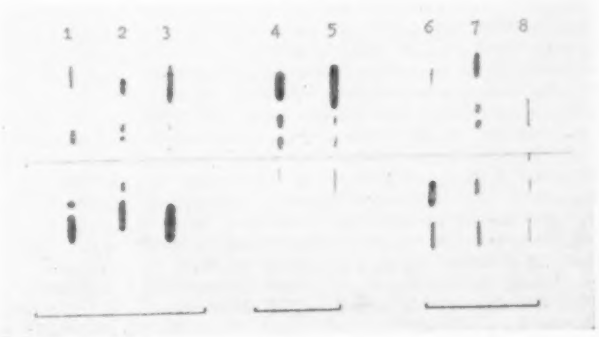

Fig. 2. Semischematic representation of zymograms. 1, Chronic ulcerative colitis; 2, adenocarcinoma of the large intestine; 3, polyp of the large intestine; 4, nodular goitre; 5, thyroid gland (disturbance in hormone synthesis); 6, bronchial carcinoma (oat-cell type); 7, bronchial carcinoma (anaplastic squamous type); 8, bronchial carcinoma (squamous type)

In principle the method employed is also suitable for other enzymes. It has already been applied by us in the case of acid phosphatase, for example.

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A Heteroglycan from *Toxoplasma gondii*

Little is known about the biochemical determinants of virulence and antigenicity in Protozoa. So far as we are aware, no immunochemical studies on *Toxoplasma gondii* have yet been reported, except the work of Hook and Faber[1]. They showed that cytoplasm-modifying and complement-fixing antibodies were related to the protein, but not carbohydrate fractions, obtained from sonically fragmented cells. Lunde and Jacobs[2] concluded that the *Toxoplasma* haemagglutination test antigen was a protein contained in the crude lysate of washed parasites; but no attempts were made to isolate and identify the soluble antigens in a relatively pure state.

During the course of our comprehensive investigations on the immunochemistry of this protozoan species, we happened to isolate and study a polysaccharide fraction with special reference to its immunobiological properties, which are described in the present communication. The *RH* strain of *T. gondii* used was obtained from Dr. Leon Jacobs, National Institutes of Health, Bethesda, Maryland, and has been maintained by serial passage in mice, using peritoneal exudate. Pure suspensions were prepared from artificially infected laboratory-bred, serologically negative Swiss albino mice, using the method of Fulton and Spooner[3], and, on an average, each yielded 2×10^7 toxoplasms. Suspensions containing 20 mgm./ml. dry weight toxoplasmata were lysed by the addition of 10 times their weight of cold (4° C.) distilled water. After centrifugation and restoration of isotonicity by addition of an equal volume of 1·7 per cent sodium chloride solution, the carbohydrate fraction was separated by fractional precipitation with cold *iso*propanol, and deproteinization was carried out with chloroform and *n*-butanol. The actual yield of polysaccharide ranged from 2·8 to 4·2 per cent, with a mean value of 3·6.

Chemical analysis showed some unusual features, the nitrogen content varying from 0·8 to 1·2 per cent, and phosphorus from 0·2 to 0·4 per cent. No lipid could be detected on degradation with dilute acetic acid. The carbohydrate fraction was readily soluble and physical homogeneity was demonstrated by electrophoretic and ultracentrifugation techniques, and the absence of protein was confirmed by low nitrogen content, lack of ultra-violet absorption in the region of 280–290 mμ and failure of products to give the usual protein reactions. No nucleic acid could be detected by examination of their ultra-violet absorption spectra.

Agar-diffusion[4] experiments demonstrated immunological homogeneity when pure polysaccharide antisera reacted with homologous fractions and proliferative cell suspensions to give only one line of precipitation. When similar fractions obtained from ten other strains of *T. gondii* were examined in the gel-diffusion test, the lines that developed indicated identical specificity since they were, in every case, contiguous when reacting from adjacent reservoirs. No reaction was evident when antiserum against *Besnoitia jellisoni* (kindly furnished by Dr. J. K. Frenkel, University of Kansas Medical Centre) was tested against these preparations, or vice versa.

Chromatographic analysis revealed the presence of glucose, glucosamine and a rare pentose which could not be identified with reagents at our disposal. The presence of these sugars was confirmed by the corresponding precipitation inhibition tests on Ouchterlony double-diffusion plates. No aldoheptoses were found, and there is suggestion that the diffuse streaks on the stained chromatograms may refer to some of the newly discovered dideoxy sugars; this point is under examination in Prof. Dr. O. Westphal's laboratory at the Dr. A. Wander Forschungsinstitut, Freiburg-Zähringen, West Germany. Tests for uronic acid and reactive acetyl gave negative results.

The polysaccharide was a poor antigen when isolated, but high-titred antisera could be produced in rabbits by combining the preparations with the lipoprotein of *Shigella dysenteriae*, or by injecting the polysaccharide after coating on erythrocytes or on killed group *A* haemolytic streptococci.

The polysaccharide sensitized sheep erythrocytes to agglutination, and lysis (120 μgm./ml.), and the specificity of the reaction was demonstrated in the corresponding inhibition tests. The fraction possessed no pyrogenic, skin-sensitizing or toxic activity, and after treatment with heat or alkali no alteration in these properties was detected. All the preparations fixed complement, did not interfere with the cytoplasm-modifying reaction, became coated on collodion particles (rendering them specifically agglutinable), inactivated the properdin system *in vitro*, had anti-phagocytic and virulence-enhancing activity and acted both as a preparing and shocking reagent in the elicitation of generalized Schwartzmann reaction in rabbits.

Studies on its topographic distribution on intact cells of *T. gondii* indicated that it is at least a partial surface antigen. Resting cell-suspensions of *T. gondii* failed to oxidize the polysaccharide, which could not deplete the dye-test or complement-fixing activity of whole-cell antisera. In all cases, the polysaccharides, derived from as many as eleven strains that were examined so far, stimulated non-specific immunity to an extent approaching that induced by *Escherichia coli*, 2380, when tested by the method of Rowley[5].

The polysaccharide specimens could neither actively immunize mice to artificial infection with *T. gondii* nor did their corresponding antisera possess any passive mouse-protective power. Besides, all the preparations were found to lack (1) Forssman heterophile reactivity; (2) direct haemagglutinating capacity; (3) capacity to inhibit lysozyme *in vitro*; and (4) ability to interfere with haemagglutination of fowl erythrocytes by Newcastle disease virus. No heterogenetic relationship could be demonstrated between these soluble antigens and many species of bacteria that were included in this work.

No serological cross-reactivity was found between polysaccharide antisera and carbohydrates of known

monosaccharide composition and linkage, such as synthetic polyglucoses with 1-4, 1-6 and 1-4-6 linkages, glycogens with 1-4-6 branching points, gum arabic and rhamnose-containing polysaccharides such as Karaya gum.

A comparative investigation is in progress to delineate the role of this antigenic fraction in infection and immunity. A detailed account of these results will be published elsewhere.

We are indebted to Dr. T. Ramakrishnan, Indian Institute of Science, Bangalore, who carried out the physical examination of the polysaccharide fraction.

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BIOLOGY

Electron Microscopy of Mossy Fibre Endings in Ammon's Horn

The endings of the axons of the granule cells of the dentate fascia in relation to Lorente de Nó's areas *CA3* and *CA4*[1] of Ammon's horn have certain special morphological characteristics as seen in Golgi preparations. These have been described by Cajal[2], who termed them mossy fibres, analogous to those of the cerebellum. He found that these axons have a tortuous course and that they show characteristic varicosities consisting of triangular or star-shaped masses of protoplasm, from the angles of which both short and long processes arise. The main course of this dentate-ammonic tract runs across the bases of the apical dendrites of *CA4* and *CA3* neurons at right angles to them. The protoplasmic masses come into intimate contact with the apical dendrite or occasionally with the neuronal perikaryon, and were considered by Cajal to be sites of synaptic contact. Lorente de Nó[1] was also of this opinion, but could not decide whether the structures were part of the pyramidal neuron or of the axon terminating on it. According to Cajal[2] these 'mossy endings' could not be stained by reduced silver techniques.

Electron microscopy shows that these structures are large presynaptic bags into which dendritic spines, sometimes branched, are invaginated.

Appropriate regions of the hippocampus of the rabbit were fixed with buffered osmium tetroxide, stained with alcoholic phosphotungstic acid and embedded in araldite for sectioning[3].

Sections through the apical dendrites near their bases show the presence of structures having the characteristics of presynaptic processes, but which have diameters of 3–4μ (Fig. 1, *pre*). These large bags are full of synaptic vesicles, and the enclosing membrane may be seen to be in close contact with the apical dendritic membrane for distances of up to 4μ. The opposed membranes are thickened at intervals (*b*), such thickenings having the appearance

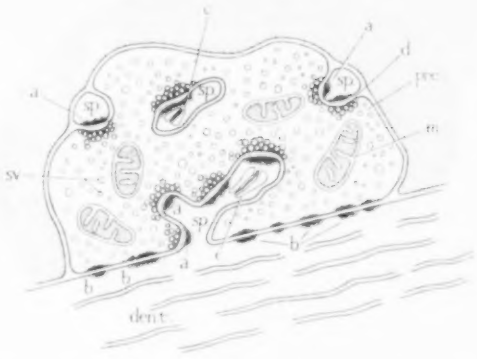

Fig. 1. Diagram of a presynaptic bag showing areas of synaptic contact with dendritic spines and with the main dendrite. The stippled regions represent neuronal and glial processes of the neuropil

of synaptic contacts as described by previous authors[3].

In addition, the profiles of several dendritic spines (*sp*) may be seen within the bag, and in favourable planes of section continuity of these with the main dendrite can be observed. Such dendritic spines may be branched. The tubules of the main dendrite (*den. t.*) do not appear to enter these spines. Identification of these small dendritic profiles (diameter is usually 0·5–1μ) depends on the presence in many cases of a spine apparatus (*c*) similar to that which has been described in neurons of the visual cortex of the rat[3]. Also, the membranes of the spine show numerous synaptic contacts (*a*), in which the thickening of the post-synaptic membrane is wider and denser than that of the presynaptic membrane, with, in addition, widening of the intermembranous space (~300 Å.) in the region of the synaptic area. These appearances are in contrast to those of the synaptic contacts between the presynaptic bag and the main dendrite (*b*), where the thickenings of the two membranes are equal and there is no increase in width of the space (~200 Å.) between the pre- and post-synaptic membranes. These two varieties of synaptic contact thus correspond to the types 1 and 2 described by Gray[3] in the visual cortex of the rat.

It is of interest that in this situation a single presynaptic bag may be observed to form two types of synaptic contact, suggesting that the differences may be due to variations in the properties of the post-synaptic processes rather than in the nature of the axonal ending. Observations of Golgi preparations[2] with the light microscope show that these synaptic structures are in many cases not terminal, but of the 'bouton de passage' type. It has not been possible so far to verify this point in electron-micrographs, owing to the difficulty of obtaining a suitable plane of section. As well as the synaptic vesicles, the presynaptic bags may be seen to contain from three to six mitochondria, but in no case have neurofilaments been observed. The absence of neurofilaments may explain the fact that these endings do not stain with reduced silver techniques, as the argentophilia which is found in the axonal terminals of certain regions appears to depend on neuro-filaments being present[4].

A full report of this work will be published at a later date.

I am indebted to Dr. E. G. Gray and Prof. J. Z. Young for their interest and advice.

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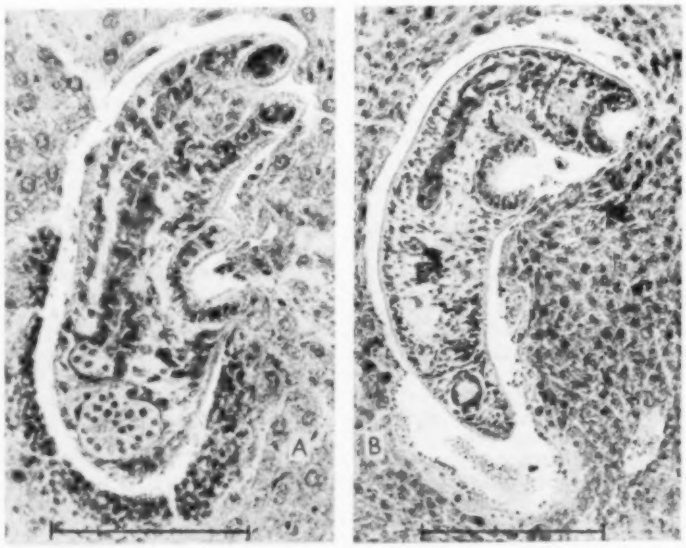
Juvenile Stages of *Fasciola hepatica* in the Liver of the Mouse

Most biologists agree that in the later stages of its life-cycle *Fasciola hepatica* wanders through the liver towards the bile ducts, where maturity is attained. When cysts containing metacercariae are ingested along with herbage to which they are attached, the larvae are activated in the intestines of the host and are then able to emerge from the cysts, each of which develops a minute perforation. The young flukes then penetrate the wall of the intestines, creep about over the abdominal viscera and eventually enter the liver. Much research and some controversy were necessary before this migration became established beyond any doubt. Schumacher[1] has reviewed the literature bearing on this problem, and himself carried out experiments of a classical nature, which produced much valuable information concerning the time relations of the various stages of the migration in guinea pigs. He also gave an account[2] of the histological appearance of young flukes in the liver of the host 64 hr. after cysts were fed, but this is a very incomplete study and practically no other information of the same kind is available in the literature of fasciolíasis. I have studied the early stages of migration through the liver of the definitive host, the mouse having been chosen to minimize the rather laborious task of serializing sections of infected liver.

The young flukes assemble in the abdominal cavity of the mouse about 24 hr. after cysts are administered *per os*, and during the next 2 days some of them enter the liver. At this stage, they are about 0·3 mm. long, but growth is so rapid that the length is about 0·5 mm. in 6 days, 0·6 mm. in 7 days, 0·8 mm. in 8 days and 1·2–1·6 mm. in 11 days. During little more than 1 week spent in the liver, therefore, the young fluke quadruples its length, at the expense of hepatic tissue. The burrow of the entering fluke is only a little broader than its body, but it is equal to about six hepatic cells in breadth, so that by the time the body of the fluke is just accommodated several hundred hepatic cells have been damaged and excavated. As the fluke grows, the burrow becomes progressively larger, and as the liver may be riddled with burrows, in even moderate infections, the toll of hepatic tissue must become truly enormous. Two photomicrographs of this damage and also the mode of progression of the young flukes are reproduced here (Fig. 1). Four days after the feeding of cysts, the young fluke may be seen advancing into a mass of hepatic cells (Fig. 1, *A*). Pyknotic nuclei of hepatic and reticulo-endothelial cells, and some cytoplasm, form a wedge-shaped mass which is entering the oral sucker. Damaged hepatic tissue is seen in the wall of the burrow and a mass of debris, largely nuclear but also containing leucocytes and erythrocytes, occurs in the wake of the fluke. This appearance is characteristic. The fluke advances into normal tissue, effectively devouring it, and broken tissue which is not ingested forms a concentrate, largely of nuclear origin, which is smoothed down by the movements of the fluke and eventually left behind in the most recently vacated part of the burrow. The burrowing fluke moves along a slightly zig-zag path, the direction of advance changing slightly from time to time, and the older burrow thus assumes the appearance of a number of ovoid chambers with narrower connexions. Change of direction is indicated by slight compression of the hepatic tissue on the right of the fluke seen in sagittal section in Fig. 1, *B*; this fluke, which is at the 8-day stage, illustrates the way in which the rim of the oral sucker, beak-like in section, bites into masses of hepatic cells, and the anterior end of the fluke is flexed ventrally in a characteristic feeding posture.

The importance of studying the feeding activities of such young flukes will be obvious to helmintholo-gists, who are well aware that such flukes are able to resist the action of any drugs which have so far proved successful for the destruction of adult flukes residing in the bile ducts. A fuller account of the early stages of migration through the liver is now published[3], and in this an attempt has been made to demonstrate the preference which young flukes seem to have for a



0·2 mm. 0·3 mm.

Fig. 1. Sagittal sections passing through two young flukes in the liver. In *A*, 4-day stage, note damaged tissue in the wall of the burrow, hepatic cells with pyknotic nuclei about to be ingested, and debris accumulating in the rear of the fluke. In *B*, 8-day stage, note a characteristic feeding posture, oral sucker approaching hepatic cells, and change of direction of burrowing indicated by compression of hepatic tissue

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diet of hepatic cells. It is inevitable that damage to a vascular organ such as the liver should produce haemorrhages, but in these early stages it is seen that much of the blood which seeps into the burrows lodges in the regions vacated by the flukes, which ever seem to seek fresh hepatic cells. It is claimed that the young fluke is never a blood-feeder in the accepted sense of the term, showing a distinct preference for the hepatic cells, which must form a succulent diet.

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Biochemical Indications as a means of distinguishing between Iron and Manganese Deficiency Symptoms in Citrus Plants

IRON and manganese deficiencies are widespread in many citrus-growing areas throughout the world, including Israel. Similarity of the visual symptoms of these deficiencies, in some stages (Fig. 1), makes differentiation difficult[1].

Fig. 1. Control (centre) and manganese- (left) and iron- (right) deficient lemon leaves

Moreover, due to the rather wide range of leaf patterns associated with manganese insufficiency, there are contradictory descriptions of these symptoms in the literature[1–3]. Since it is assumed that the visible symptoms are, in some way, an expression of the disturbed metabolic processes in plant cells, it was decided to investigate these metabolic pathways in order to throw some light on the reasons for this visual similarity. Different metabolic patterns, however, may be used as reliable indicators in differentiating between iron and manganese deficiency symptoms.

This communication deals with only a part of the different metabolic patterns that were observed.

For the purpose of the experiment, several varieties of 3-month-old citrus seedlings—rough lemon, sweet lime and sour orange (generally used as root-stocks) and Shamouti orange, Duncan grape fruit, and Eureka lemon (used as scions)—were transferred to aerated water culture and were grown in a greenhouse. Purification and composition of the nutrients were according to Hewitt[4].

The solutions contained gradually decreasing amounts of iron and manganese and were changed every fortnight. Characteristic deficiency symptoms developed after 3–6 months. Leaf samples were taken from the plants exhibiting different stages of the developing visual symptoms, especially when iron and manganese deficiencies could be confused.

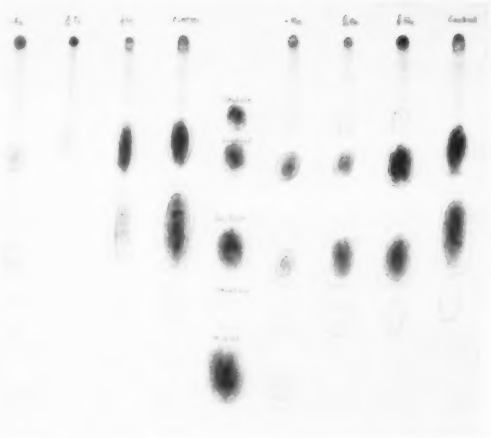

Fig. 2. Ascending paper chromatograph (butanol/acetic acid/water, 9 : 1 : 3) of lemon leaves extract from plants supplied with decreasing amounts of iron and manganese, compared with the markers (in the centre). Spots developed by aniline/butanol/phosphoric acid/water (1 : 5 : 1·2 : 10) spray

Five gm. of fresh leaf material were macerated in 80 per cent ethanol, the ethanol was evaporated *in vacuo*, and the solution was passed through 'Zeokarb 225' and 'Deacidite *G*' resin columns which retained amino-acids and organic acids respectively. The solution, after passage, contained sugars which were detected by means of paper chromatography[5].

Fig. 2 clearly shows the degradation of sugars, due to decreasing amounts of iron in the nutrient solution, and the appearance of some free pentose in the case of manganese-deficient lemon plants.

Since pentoses can be more rapidly detected by methods other than chromatography, it was decided to do so by means of an orcinol test, which can be used as a simple colorimetric determination[5].

For this purpose 0·5 gm. of fresh leaf material was macerated in water and acetone, and besides the pentoses, the peroxidase activity and the chlorophyll content of the sample were estimated[6]. Chlorophyll serves as a means of indicating degree of chlorosis, and peroxidase activity has been found to vary, according to different micronutrient deficiencies, in tomato plants[7].

After centrifugation the supernatant was purified by active carbon and 'Zeokarb 225', and the colourless plant sap was used for the orcinol reaction[5].

Table 1 shows that, in addition to free pentose accumulation, which is enhanced by manganese deficiency, peroxidase activity and chlorophyll $a : b$ ratio may give some substantial contribution to the problem of rapid and correct evaluation of this type of deficiency symptom. This 'biochemical approach' should be appraised in the light of other diagnostic methods, namely, visual symptoms, mineral analysis of the leaves and different methods of diagnostic supply of minerals to deficient leaves.

Chemical analyses of leaves for iron and manganese are not only troublesome in comparison with these tests, but also do not always give a conclusive answer; for example, it has been found[2] that when manganese is deficient, in many cases, less iron also appears to accumulate in the leaves. Diagnostic sprays consume considerable time and, sometimes due to impurities

Table 1. Effect of Manganese and Iron Deficiencies on Chlorophyll, Pentose Content and Peroxidase Activity of Citrus Leaves*

Citrus variety	Treatment	Chlorophyll (percentage of fresh material) Total	*a*	*b*	*a* : *b*	Peroxidase activity†	Free pentose content‡
Rough lemon	Control	0·1040	0·0770	0·0270	2·84	33	100
	−Mn	0·0507	0·0332	0·0175	1·89	12	145
	−Fe	0·0501	0·0364	0·0137	2·64	180	85
Sweet lime	Control	0·120	0·0843	0·0356	2·40	27	100
	−Mn	0·0709	0·0459	0·0250	1·83	13	133
	−Fe	0·0411	0·0288	0·0123	2·34	61	68
Sour orange	Control	0·0954	0·0680	0·0274	2·48	47	100
	−Mn	0·0852	0·0540	0·0312	1·70	14	147
	−Fe	0·0477	0·0326	0·0151	2·16	73	92
Eureka lemon	Control	0·0822	0·0596	0·0226	2·63	32	100
	−Mn	0·0339	0·0223	0·0116	1·92	15	222
	−Fe	0·0362	0·0268	0·0094	2·85	59	109
Duncan grapefruit	Control	0·0849	0·0615	0·0234	2·62	32	100
	−Mn	0·0687	0·0452	0·0235	1·92	16	194
	−Fe	0·0231	0·0154	0·0077	2·13	125	88
Shamouti orange	Control	0·0951	0·0668	0·0283	2·36	48	100
	−Mn	0·0645	0·0423	0·0222	1·93	21	180
	−Fe	0·0212	0·0152	0·0060	2·51	154	114

* Each value is an average of three or four leaf samples of different ages.

† Peroxidase activity is expressed as number of seconds which were required to complete oxidation of 75 μgm. pyrogallol at *p*H by 5 mgm. fresh leaf material.

‡ Each leaf sample, even the control, developed some colour with the orcinol reagent. The value of the control has been taken arbitrarily as 100.

in the chemical compounds used in these sprays and injections, the results are open to doubt. This 'biochemical approach', therefore, may serve as an additional means of diagnosis in cases of uncertainty.

This work was carried out at the Hebrew University under the supervision of Prof. H. R. Oppenheimer.

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Oxidation of Reduced Pyridine Nucleotides and of Sulphite by Pea Root Mitochondria

It has been shown[1] that freshly isolated pea root mitochondria oxidize α-oxoglutarate poorly or not at all if the oxygen uptake is measured by a polarographic method (in which the mitochondria are not shaken), and the oxygen uptake is not stimulated by diphosphopyridine nucleotide (oxidized form, DPN). However, if the mitochondria are first shaken for 30 min. in a water bath at 25°, they do oxidize α-oxoglutarate and DPN increases the rate of oxygen uptake (Table 1).

The ability of these mitochondrial preparations to oxidize the reduced pyridine nucleotides has been examined by the same procedures. At a low concentration (0·1 m*M*) reduced diphosphopyridine nucleotide (DPNH) was not oxidized by unshaken, but was by shaken, mitochondria (Table 2, exp. 1). This suggests that shaking makes the mitochondrial membrane more permeable to DPNH. With reduced

Table 1. Oxidation of α-Oxoglutarate

Additions	Q_{O_2} Unshaken mitochondria	Q_{O_2} Shaken mitochondria
None	450	350
DPN (0·1 m*M*)	460	350
α-Oxoglutarate (10 m*M*)	440	660
DPN (0·1 m*M*) + α-oxoglutarate (10 m*M*)	440	790

The incubation medium contained 0·05 *M* phosphate buffer, *p*H 6·8, 10 μ*M* cytochrome *c*, 100 μl. mitochondrial suspension and additions as given in the Tables, in a total volume of 0·5 ml. Experiments were conducted at room temperature (22–23° C.). Oxygen uptake (measured polarographically) is expressed as Q_{O_2} (μl. O_2/mgm. trichloroacetic acid—insoluble N/hr.).

triphosphopyridine nucleotide (TPNH), however, the oxygen uptakes were about the same for both unshaken and shaken mitochondria (Table 2, exp. 2), thus indicating, perhaps, that the membrane of the unshaken mitochondrion is already permeable to TPNH (but not to DPNH). In this respect pea root mitochondria are similar to rat liver mitochondria[2].

Table 2. Oxidation of Reduced Pyridine Nucleotides

Exp.	Additions	Q_{O_2} Unshaken mitochondria	Q_{O_2} Shaken mitochondria
1	None	500	500
	DPNH (0·1 m*M*)	510	740
2	None	530	560
	TPNH (0·1 m*M*)	630	680
	TPNH (0·7 m*M*)	740	780
3	None	520	490
	DPNH (5 m*M*)	2,110	1,610
	TPNH (5 m*M*)	1,430	830

At much higher concentrations (5 m*M*), both DPNH and TPNH were oxidized by unshaken mitochondria (Table 2, exp. 3). They were also oxidized by shaken mitochondria, but at a much slower rate. At a concentration of 1·0 m*M*, DPN enhanced the low oxidation of α-oxoglutarate by unshaken mitochondria[1] whereas there was no enhancement at a concentration of 0·1 m*M* (Table 1). It thus seems that at external concentrations of 1·0 m*M* and more, the nucleotides can enter the mitochondria.

Table 3. Oxidation of Sulphite

Exp.	Additions	Q_{O_2} Unshaken mitochondria	Q_{O_2} Shaken mitochondria
1	None	500	430
	ADP (m*M*)	510	440
	Na_2SO_3 (0·3 m*M*)	500	580
	ADP (m*M*) + Na_2SO_3 (0·3 m*M*)	500	690
2	None	400	410
	DPN (0·1 m*M*)	400	400
	Na_2SO_3 (0·3 m*M*)	410	560
	DPN (0·1 m*M*) + Na_2SO_3 (0·3 m*M*)	420	560

Sulphite is oxidized by mitochondrial preparations from pea internodes[3] and oat seedlings[4]. It is also oxidized by pea root mitochondria, but only if these were first shaken (Table 3). Adenosine diphosphate (ADP) increased the uptake of oxygen in the presence of sulphite only with shaken mitochondria and DPN

had no effect on this uptake. This supports the finding[3,4] that ADP, but not DPN, is required by the sulphite-oxidizing system.

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Growth of *Fomes annosus* under Reduced Oxygen Pressure and the Effect of Carbon Dioxide

Although most fungi are regarded as strictly aerobic organisms, some species—under natural conditions—are known to colonize in localities which *a priori* must be expected to offer only poor supplies of free oxygen. This is the case with fungi attacking the interior of roots and stems of living trees. Owing to respiration in the living tissues of these organs, free oxygen dissolved in the water of vessels in sapwood and heartwood or present as free gas in intercellular spaces and non-functioning vessels is soon consumed. Concomitantly carbon dioxide is liberated. Diffusion of gases within a tree trunk probably occurs by the shortest distance, namely, by horizontal diffusion from the outside through the cortex into the interior of the stem and vice versa. However, in warm seasons when respiration-rate is high the very active cambium enclosing the central wood of a stem offers a barrier to the diffusion of gases in either direction[1]. Consequently as oxygen is consumed by living cells (medullary ray cells, parenchyma cells) in the sapwood the oxygen pressure will drop rapidly in the interior and be kept at a low level until diffusion is again made possible by a decrease in temperature.

Seasonal variation in oxygen and carbon dioxide pressures in stems of several hardwood and softwood trees has been investigated by Chase[2], who found that in summer the oxygen content in the wood of *Pinus strobus* would drop to approximately 1 per cent, corresponding to a pressure of 8 mm. mercury. At the same time the carbon dioxide content increased to approximately 10 per cent.

The parasitic basidiomycete *Fomes annosus* (Fr.) Cke. is known to attack roots and heartwood of many conifers and even deciduous trees, thus being exposed periodically to conditions of low oxygen pressure and high carbon dioxide pressure. It was, therefore, of interest to investigate the growth response of this fungus to such conditions.

Round 6-mm. pieces from the periphery of a young colony of *F. annosus* on malt agar were inoculated on to the surface of freshly boiled and poured malt agar in Petri dishes and incubated at room temperature in Brewer jars containing air, hydrogen, nitrogen and/or carbon dioxide. In the hydrogen jars all free oxygen and oxygen dissolved in the agar was removed by catalytic reaction with the hydrogen, the jars thus representing a strict anaerobic environment. No growth occurred in these jars (Table 1). In the nitrogen jars the oxygen was removed by three subsequent evacuations to 40 mm. mercury and slow refillings with nitrogen which had passed through a wash bottle containing alcalic pyrogallol. Theoretically the nitrogen jars contained only 0·02 mm. oxygen

Table 1. Growth of *Fomes annosus* on Malt Agar in Air, Nitrogen, Hydrogen and Carbon Dioxide. Total Pressure 760 mm. Mercury. Incubation 6 days at Room Temperature

Gas pressure (mm. mercury)				Linear growth (mm.)	
O_2	N_2	H_2	CO_2		
160	600	0	0·228	30	Atmospheric air
155	582	0	23	44	Carbon dioxide added to air
0	trace	760	(trace)	0	
0	trace	737	23	0	
trace	760	0	(trace)	30	
trace	737	0	23	45	

although the actual content might have been somewhat higher because of sluggishness in oxygen diffusion from the agar. Nevertheless, these bottles represented a micro-aerophilic environment in which *F. annosus* proved to grow unaffected.

For growth in pure carbon dioxide the jars were filled in the same way as with nitrogen except that the washing in pyrogallol was omitted. These jars also represented a micro-aerophilic milieu. With 760 mm. carbon dioxide no growth occurred whereas 23 mm. of the gas in combination with air or nitrogen accelerated growth about 50 per cent. As could be expected, no effect was obtained under strictly anaerobic conditions in hydrogen.

That neither hydrogen nor carbon dioxide in high concentration *per se* was toxic to *F. annosus* was shown by subsequent aerobic incubation at which normal growth was resumed.

Nine different strains of *F. annosus* (isolated from fruiting bodies on infected *Pinus sylvestris* and *Picea abies*) have responded qualitatively equal to these conditions. It could be concluded that *F. annosus* is a fungus able to grow equally well under aerobic as under microaerophilic conditions, but not anaerobically, and that a slight elevation of the carbon dioxide pressure has a stimulatory effect, but a high concentration of this gas an inhibitory effect on mycelial growth. These observations seem to agree well with what was to be expected from a fungus parasitizing the interior of roots and trunks.

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Filtration of Phytoplankton by the Octocoral *Alcyonium digitatum* L.

Most of the work carried out on the food and feeding of coelenterates deals with the true reef-building corals, Actiniae and Scyphomedusae, which harbour zooxanthellae in their tissues. The group Alcyonaria, however, has received less attention. There seems to be considerable controversy as concerns the significance of these symbiotic algae for their host-coelenterates. Some authors report that the coelenterates derive no benefit from their zooxanthellae[1,2], while others believe that zooxanthellae form a considerable part of their food, the other part being of animal origin[3–5]. Still others maintain that some coelenterates, namely the xeniid octocorals, may in some way or other rely on their algae as their only source of food[6,7].

Little work has so far been done to study the food and feeding of alcyonarians which do not harbour zooxanthellae in their tissues, for example, *Alcyonium digitatum* L.[8,9], which has been described as carniv-

orous. Its food is reported to consist solely of other small animals, mainly zooplankton organisms, which it paralyses by the action of its nematocysts.

In order to investigate whether *A. digitatum* is fully dependent on animal food or may also feed on phytoplankton organisms, fresh colonies were placed in the dark in jars containing membrane-filtered sea water, to which strains of *Sceletonema costatum* labelled with carbon-14 were added. Two different concentrations of algae were used (20 : 1). The rate of removal of algae by the octocoral was estimated by measuring the radioactivity in sub-samples of 10 ml. water at time-intervals starting with half an hour. As a control, the drop in the radioactivity of the algae was measured in the same way in identical concentrations of *Sceletonema*. The considerable decrease (27 per cent) of activity in the control with the higher concentration of algae is probably due to the dense concentration of algae as well as to the bacterial activity (the *Sceletonema* strains used were not bacteria-free) (see Table 1).

Table 1. Radioactivity (c./min.) in sub-samples of 10 ml. sea-water

Lapse of time (hr. and min.)	Experimental (*A. digitatum*)		Control	
	Concentration of algae (20)	(1)	Concentration of algae (20)	(1)
0	687	45	666	34
0·30	—	34	—	—
1	674	—	—	—
2	—	17	—	—
3	533	25	—	29
4	—	65	—	—
5	492	—	592	32
6·30	496	23	—	—
23	250	—	486	36

In the higher concentration of algae, one colony of *A. digitatum* filtered about 50 per cent of the *Sceletonema* present in 23 hr. while in the lower concentration, another filtered up to 93 per cent of the algae in only 6½ hr. After the experiments, thoroughly washed pieces of the *Alcyonium* colonies were homogenized and the activities of the different parts measured (c./min.) were as follows: ectodermal covering, 4·7; mesogloeal and endodermal tissue, 22·7; and gastric juice, 46·5. In some other experiments still higher activities have been measured in the tissue-homogenates of *A. digitatum*[9]. In all the feeding experiments we have carried out using strains of *Sceletonema costatum*, *Ditylum* sp. or natural phytoplankton populations labelled with either carbon-14 or phosphorus-32, *A. digitatum* has proved to be able to filter such algal organisms.

The ability of *A. digitatum* to filter and obviously digest phytoplankton algae is evident from the above data, demonstrating the transportation of the labelled nutriment to different tissues of the colonial animal. This phenomenon may help to explain why this species can survive for relatively long periods of time in waters deficient in zooplankton, and moreover supports the idea that at least some coelenterates may also feed on plant material.

The ciliary movements in *Alcyonaria* seem well suited to filter phytoplankton algae out of the surrounding water. In *A. digitatum* the ciliary current maintained in and out of the polyps seems not only to convey oxygen, as already stated[10], but will also constantly bring a new potential source of plant food within their reach. It might, of course, be expected that such a current would play no part in providing *A. digitatum* with its animal-food, since the latter mainly consists of small zooplankton crustaceans which move too fast to be caught by ciliary currents. In the active stage, however, *A. digitatum* expands greatly with a resulting enormous increase in the surface area[11] and this brings the colony in contact with a still larger volume of water masses. Moreover, as may also be expected, expansion and extension of the polyps activate the ciliary currents in their coelentera. It is not believed, however, that expansion of the *Alcyonium* colony to its normal activity size is induced by any need of oxygen, provided that this gas is not deficient in the surrounding water[11]. It is almost certain that the expansion of the colony in Nature is related mainly to feeding requirements.

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VIROLOGY

Inhibition by Cortisone of the Synthesis and Action of Interferon

The administration of cortisone and certain other C-21 and 19-nor-steroids increases final yields of influenza virus in chick embryos by inhibition of viral interference[1]. Paradoxically, the rate of viral synthesis is reduced[2] at the same time. Interferon, a substance produced by cells inoculated with interfering virus, has been proposed as the mediator of interference[3]. Therefore, it was important to determine whether cortisone and other steroids capable of inhibiting the induction of interference might influence either the production of interferon or its effect.

We find that interferon is produced most easily by a method suggested by Wagner[4]; this involves the intra-allantoic inoculation of chick embryos with small amounts of infective virus and prolonged (65–72 hr.) incubation before harvesting allantoic fluid. After dialysis of allantoic fluid at *p*H 2·0, which destroys the virus but leaves interfering (interferon) activity, this activity is measured as the reduction in viral synthesis *in vitro* in chorioallantoic membrane cultures. It has been found that membranes of older (12–13 day) chick embryos are more susceptible to the interferon effect[5], and these have been used in the present experiments.

In Table 1 we present the summary of an experiment in which cortisone (diethylaminoacetate hydrochloride of hydrocortisone) in two dosages clearly prevented the *in ovo* synthesis of interferon. In this

Table 1. Inhibition of Interferon Formation by Cortisone

(*A*) Production of interferon (*in ovo*)

Group	−2 hr.	0 hr.	+70 hr.
1	Water	virus*	harvest and dialyse at *p*H 2·0†
2	*SC*-5914 (1·0 mgm.)	virus	
3	Cortisone (0·1 mgm.)	virus	
4	Cortisone (1·0 mgm.)	virus	
5	Water	saline	
6	—		

(*B*) Test for interferon (*in vitro*)

Group	−20 hr.	0 hr.	Haemagglutinin yield at 42 hr.
1	Incubate 1 : 2 dilution in medium with *CAM*‡	add challenge virus§	1·28
2			2·3
3			4·7
4			5·2
5			4·7
6	(Control)		5·5

* Influenza *A* (*PR8*) 10^7 EID_{50}.

† Then readjusted to *p*H 7·0; note that dialysis removes steroids from fluids.

‡ Influenza *B* (Lee) 10^6 EID_{50}.

§ Log_2 viral haemagglutinin.

‖ Chorioallantoic membrane tissue culture.

experiment 11-day-old chick embryos (4/group) were inoculated with $10^{2\cdot0}$ EID_{50} of the *PR8* strain of influenza *A* virus, or a saline control 2 hr. after the injection of water (control), cortisone, or *SC*-5914, 'Searle' (17-ethyl-19-nortestosterone), a steroid with 1/1,000 the activity (by weight) of cortisone in negating interference with inactivated virus (Kilbourne, E. D., unpublished results). After 70 hr. of incubation, allantoic fluid pools were collected from each group, then dialysed against 0·1 *M* sodium chloride glycine buffer at *p*H 2·0 for 24 hr. at 4° C. These 'interferon' preparations were then restored to *p*H 7·0 by dialysis against 0·01 *M* phosphate buffered saline for 24 hr. (two changes) (Table 1, part *A*).

As shown in part *B* of Table 1, the interferon activity of 1 : 2 dilutions of the pools thus treated was determined *in vitro* by inoculation of 12-day-old chorioallantoic membrane cultures with 10^6 EID_{50} of influenza *B* virus (Lee) 20 hr. after their incubation with 'interferon' pools or medium alone (group 6). Measurement of yields of haemagglutinin 42 hr. later demonstrated reduced titres (evidence of interferon action) only in groups 1 and 2, which had not received cortisone originally. It has been established since then that cortisone does not merely prevent the release of interferon from the chorioallantoic membrane into the allantoic fluid, but also inhibits its formation in the membrane. The design of the foregoing experiment precludes the possibility that cortisone acted *in vitro* after interferon was synthesized, because dialysis separates effectively the (dialysable) steroid remaining in the allantoic fluid from the non-dialysable interferon.

However, it has been shown in other experiments that cortisone and other steroids administered *in vitro* will prevent the *effect* of pre-formed interferon if they are given 2 hr. before the inoculation of interferon, then removed by washing before the incubation of membranes with interferon and the subsequent inoculation of challenge virus. It is of interest that suppression of interferon action is the only biological action manifestly common to the disparate steroids capable of inhibiting interference.

Previous papers from this laboratory have suggested that inhibition of both viral synthesis and interference by cortisone might have a single biochemical basis[1], and that the basis was most probably related to the inhibiting effect of cortisone on protein synthesis[2]. It is, therefore, of great interest to find that cortisone inhibits the synthesis of a protein (interferon) which can interfere with virus replication. If interferon is an abortive product of viral multiplication[3], then further evidence is provided that the effects of cortisone on viral multiplication are on protein rather than nucleic acid synthesis.

It seems possible that endogenous adrenal corticoids may influence the dynamics and outcome of infection *in vivo*, depending on the balance of their effects on viral synthesis and on the synthesis and action of interferon.

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Apparent Release of Tobacco Mosaic Virus in Living Infected Cells

Using a Zeiss *GFL*65 phase microscope with 'Neofluar' objectives we are attempting to record the sequence of changes produced by tobacco mosaic in living cells of Turkish tobacco. Leaves at intervals after inoculation are prepared by exhausting them of air under water with a high-vacuum pump. Hand sections are cut in a pan of water under a binocular microscope with small pieces of razor blade mounted in a handle. Most sections are paradermal, along or beside veins on the underside of leaves inoculated with tobacco mosaic virus on the upper surface, or on systemically infected leaves. One cell thickness, sometimes obtainable, is ideal; 2 cells may allow images that can be photographed, and occasional observations are possible on pieces 3 cells thick.

A number of tissue pieces have provided images, apparently of the primary emission of virus in recently infected cells. Strains *U*1 and *U*5 [1,2], representing 2 forms of the virus[2], initiated similar series of events, differing in several details. Appreciation of these events, when they were first seen to occur, was aided by previous studies on the state of cells after infection was apparent.

Tobacco mosaic virus aggregates in recently infected cells often appear as translucent monolayers in the parietal cytoplasm. Single or double layers are termed grey plates[4]. The long axes of the virus particles seem to be vertical to the cell wall, and aggregation is lateral. The grey plates vary in size from 1 to 5μ or more in diameter. If the virus is of the *U*1 form, several monolayers may appear over each other, remain attached, and become by accretion the familiar hexagonal crystals or thick virus plates. Virus of the *U*2–*U*5 form fails to aggregate into crystals, apparently because the end-to-end bonding of the particles is too weak to hold a number of monolayers together. Later, the virus particles of *U*2–*U*5 combine in crude acicular aggregates. A second difference between forms is the presence of

x-bodies in *U1*-infected cells, but not in *U2–U5*-infected cells.

A condition characteristic of most active plant cells, but particularly noticeable in cells in which the virus is about to appear, is the non-random distribution of organelles in the cytoplasm. The most obvious example is an apparently functional association between nucleus and plastids, in which specialized structures are involved. Chloroplasts become tightly adherent to the nucleus and remain so in spite of rapid streaming of cytoplasm around them. Approaching the nucleus before adhering to it, a chloroplast may put out a fine protrusion that attains a length of 5–30μ. The protrusion touches the nucleus and shortens as the plastid comes closer. A second protrusion from the opposite side of the same chloroplast occasionally appears, and becomes surrounded by spherosomes, mitochondria, etc. When the plastid itself becomes adherent to the nucleus the first protrusion may be still visible, extending over the surface of the nucleus; the second may have retracted. More than 30 chloroplasts have been seen adherent to a large nucleus.

The first sequence of virus emission seen and photographed was in an epidermal cell from the underside of a leaf inoculated 3 days earlier with *U5*. The microscope was focused on the nucleus of a cell, free from any indication of virus infection, between two obviously infected cells. A number of plastids adhered to the nucleus, and plastid projections were visible on its surface. The cytoplasm flowed actively over the nucleus. After half an hour, bubbles were seen around the nucleus, and bubbling continued actively for 5–10 min. Similar bubbling has been induced experimentally by physico-chemical agents (unpublished work) and seems to indicate a radical change in permeability of the nuclear membrane and nearby endoplasmic reticulum. Mitochondria and spherosomes were present and circulated in the adjacent cytoplasm. After the bubbling, plastids were no longer closely adherent to the nucleus, and gradually they became free to drift away in cytoplasmic streams. Cytoplasmic flow from the nucleus became concentrated mainly in one direction. The outflowing cytoplasm was denser than normal, and monolayer grey plates with a dark 2- or 3-layer spot in the centre began to appear (Fig. 1). A slower rate of flow from the other side of the nucleus allowed condensation of a few grey plates on the nuclear surface. The source of virus material seemed to be inside the nucleus, possibly around the nucleolus. In several cells when increase of visible virus ceased, the nucleolus was noticed to have lost its optical density or seemed smaller than it was before virus began to appear. Active flow of virus material from the nucleus continued for 10–15 min. (longer in other examples), and the grey plates circulated through the cell, giving it the typical appearance of a recent infection.

Fig. 1. Late stage in virus emission, virus aggregates (grey plates) formed and at a distance from the nucleus. Flow from the nucleus still apparent

The second full sequence seen and photographed was with strain *U1* in a sub-epidermal cell from a young systemically infected leaf 4 days after inoculation. The preliminary bubbling around the nucleus was less active than in the *U5* series, but in this and other examples of *U1* infection there seemed to be a second site of activity in the cell. From around the nucleus with its adherent plastids, rapid and copious streaming occurred towards a vortex in the cytoplasm, some distance away. The basis of the vortex was a complex of cytoplasmic vesicles. Such vesicles have been interpreted as inclusions of vacuolar material within the cytoplasm[1]. In the vortex, spherosomes and mitochondria were in violent motion. Virus appeared as grey plates or as less-formal aggregates, mainly in the sluggish return flow from the vortex to the nucleus. The equivalent of a cytoplasmic vortex in the *U5* infections seemed to be near or around the nucleus itself, rather than at a distance, as in *U1*.

In several *U1*-infected cells, seen after emission, amorphous material was present against the nucleus among the plastids. It drifted away with some of the plastids and circulated, apparently as the beginning of an *x*-body. It should be possible from further observation and biochemical tests to determine whether this material contains any virus protein or nucleic acid, or is debris from cell structures such as plastids.

The whole appearance of these events suggests a burst of virus emission quickly accomplished, rather than a mere assembly of virus on or at various distances from the nucleus. If the events have not been wrongly interpreted, they indicate that the final stages of tobacco mosaic virus multiplication begin within the nucleus. This agrees with Zech's findings[5]. The nucleolus seems to lose material during virus emission, and may be the source of virus nucleic acid. Possibly plastids are concerned with the synthesis of virus protein[6], and in the cytoplasmic vortex most of the nucleic acid and protein may be joined together. Finally, aggregation into grey plates, or other forms, often seems associated with vesicles or vacuolar membranes. Although such suggestions about the steps in virus formation are almost entirely speculative, it can be claimed with reason that whole virus is formed very soon after the burst of activity initiated in and around the nucleus of recently infected cells.

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GENETICS

Frequencies of Hairy Pinnæ among Indian and Sinhalese Peoples

SINCE Tommasi[1,2] published a small pedigree, one of the phenotypes suspected to be holandrically inherited in human beings is that called hypertrichosis of the ears but more accurately hypertrichosis of the pinnæ of the ears. I have shown this to be completely Y-linked in my family, publishing a pedigree containing 181 people of 7 generations[3]. The age of onset is usually in late puberty, though in a few cases the hairs are not conspicuous until the men are about 25.

I have examined 495 men in India, and 414 men in Ceylon to count the frequency of this characteristic. These figures are given in Table 1. Andhra Pradesh is a south-eastern State of India with a population of about 33,000,000. In this State I counted the frequency of this characteristic among my co-travellers in buses and railway trains during 11 journeys travelling a distance of about 800 miles[3]. In the different parts of Ceylon where the counts were made many subjects stated that their ancestors had been local people for a long time.

Table 1

Place	Subjects	Total No.	No. with hairy pinnæ	Percentage with hairy pinnæ
India				
Andhra Pradesh	Co-travellers	345	21	6·1
West Bengal	Workers of Indian Statistical Institute	150	24	16·0
Ceylon				
General Hospital, Colombo	Outpatients	124	44	35·5
Coconut Research Institute, Lunuwila	Staff and labourers	109	35	32·1
Coconut Seed Multiplication Farm, Ratmalagara	Staff and labourers	60	26	43·3
Department of Agriculture, Peradeniya	Workshop men	58	23	39·7
Royal Botanic Gardens, Peradeniya	Garden staff	63	25	39·7

Statistical analysis shows that : (*a*) there is no significant heterogeneity between frequencies in different parts of Ceylon ; (*b*) the frequencies of samples in Ceylon, Andhra Pradesh, and West Bengal differ significantly. Among the Sinhalese 295 were Buddhists most of whose ancestors had probably lived in Ceylon for 2,000 years ; 97 were Christians, mostly Roman Catholics ; 12 Muslims and 10 Hindus. Three of the Muslims and 3 of the Hindus had hairy pinnæ. There is no significant difference between the frequencies among Buddhists (37·8 per cent) and Christians (39·2 per cent).

I have also examined many women in the various localities but found none having hairy pinnæ, as is expected on the hypothesis of Y-linked inheritance. The fact that the frequency of hypertrichosis is higher in Bengal and Ceylon than in an intermediate region is compatible with the statement in the Mahawamsa, an ancient Sinhalese chronicle, that Ceylon was colonized from Bengal about 2,500 years ago.

The results presented here clearly show significant differences in the frequency of the hairy pinnæ between different populations. It would seem worth while making counts of the frequencies in various parts of the world. This requires very little effort and may prove to be as useful as the survey of blood groups and other biochemical characteristics in obtaining confirmatory evidence about the affinities and differences between different human populations.

I am grateful to all the subjects for their co-operation, and to the following people in Ceylon for helping in the survey : Drs. W. Pallie, V. Basnayake, M. L. M. Salgado, I. Samaraweera, J. Wanigasooriya and Mr. W. A. K. Fonseka.

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Karyological Demonstration of Hybridization of Mammalian Cells *in vitro*

IN a recent publication, Barski, Sorieul and Cornefert[1] described the formation of cells with a hybrid karyotype in mixed cultures of two mouse cell strains *in vitro*. The experiments to be described show that this phenomenon is easily reproducible and that it is not limited to a pair of strains of strictly defined karyotypes.

The two cell lines used in our experiments are strains *NCTC* 2555 and *NCTC* 2472 of Sanford, Likely and Earle[2], ultimately derived from a single cell of mouse subcutaneous adipose tissue, as received from Dr. G. Barski by kind permission of Drs. Wilton R. Earle and Katherine K. Sanford. Both strains are populations composed predominantly of hypotetraploid cells but containing varying proportions of cells of higher ploidy[3]. The karyotypes of the hypotetraploid cells of the two strains, as established in the course of our experiments, are given in Table 1 and illustrated by Figs. 1.*A* and *C*.

Table 1

	NCTC 2555	*NCTC* 2472
Total number of chromosomes, modal	57	55
Total number of chromosomes, variation extremes	51–64	51–59
Number of biarmed chromosomes, modal	15	1
Number of biarmed chromosomes, variation extremes	11–16	1–3
Number of extra long telocentrics, modal	0	1
Number of extra long telocentrics, variation extremes	0	1–3
Minutes	?	0

It can be seen that the two cell types can be unmistakably identified by the presence of (*a*) a high number of bi-armed chromosomes, some of which represent good 'markers', in the cells of line *NCTC* 2555 ; (*b*) at least one extra long chromosome and a somewhat shorter one (not shown in Table 1, but visible in Fig. 1,*C*) with a sub-median secondary construction in the cells of line *NCTC* 2472.

Mixed cultures were initiated on January 3, 1961, by inoculating equal numbers of cells of the two lines into two media (a modified 199 (ref. 4) and *NCTC* 109 (ref. 5)) containing 10 per cent of heat-inactivated horse serum. Dispersion of cells for transfer was accomplished either mechanically or by trypsinization. The cultures were incubated at 37° C. in an atmosphere of 5 per cent carbon dioxide in air. Control cultures of the two strains separately were treated

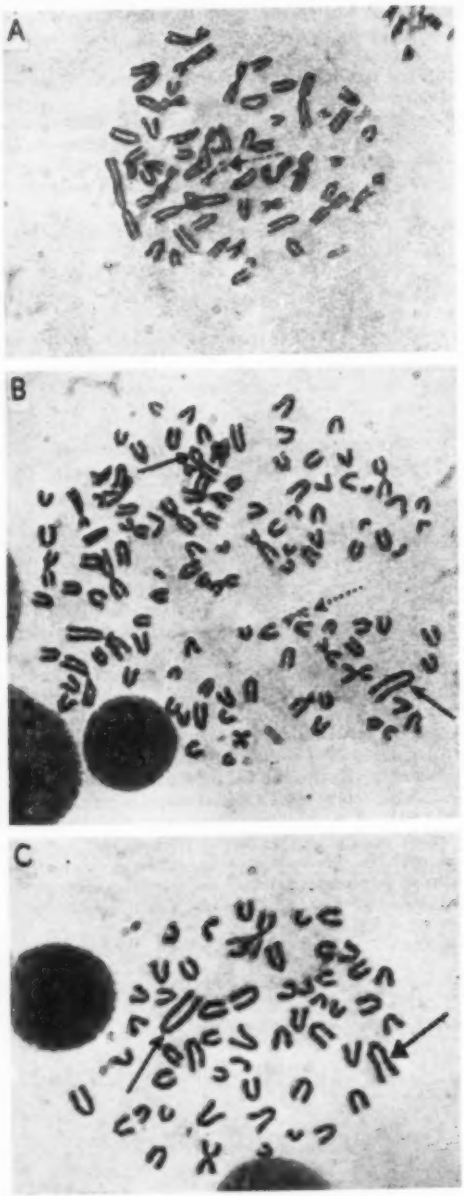

Fig. 1. *A*. Metaphase with 58 chromosomes (of which 15 are bi-armed) in a cell of strain *NCTC* 2555. Note characteristic metacentric (arrow) present in almost all cells of the strain. *B*. Representative example of hybrid cell with 112 or 113 chromosomes, of which 16 or 17 are bi-armed. Note the presence of the three 'markers' (arrows) shown in *A* and *B*. *C*. Metaphase with 54 chromosomes (2 bi-armed) in a cell of strain *NCTC* 2472. Note the two 'marker' telocentrics: the extra long one and the somewhat shorter, submedially constricted one (arrows)

similarly. Karyological analyses of the mixed populations and of cells of each line grown separately were carried out at intervals on aceto-orcein squashes of colchicine-treated cultures.

At the time of writing (March 28), the occurrence of hybrid cells was observed in all four of the karyologically analysed mixed populations (two in medium 109 and two in medium 199). The hybrid cells are recognized by the total number of chromosomes and the number of bi-armed chromosomes expected from the fusion of hypotetraploid cells of the two strains, as well as by the presence of 'marker' chromosomes (Fig. 1.*B*). The frequencies of hybrid cells in the four mixed populations are at present approximately 1, 1, 5 and 10 per cent, the highest value having been observed in medium 109.

These results confirm the observations of Barski *et al.*, and furthermore show that the formation of hybrid cells is easily reproducible and is not restricted to two rigorously defined karyotypes: the latter is clear from the fact that strain *NCTC* 2472 used by us and strain *N* 1 *T* (derived from *NCTC* 2472) used by Barski *et al.* have somewhat different karyotypes. Thus the long constricted chromosome of line *NCTC* 2472 used by us has never been observed in Barski's line *N* 1 *T*; the cells of the latter strain very seldom contain bi-armed chromosomes, whereas those of line *NCTC* 2472 used by us all carry at least one such chromosome.

It may be pointed out also that, of the two strains involved in the mixed cultures of Barski *et al.*, at least one was re-isolated from a tumour induced by the inoculation of the original *in vitro* line. On the other hand, the cells used in our experiments never underwent animal passage since their explantation by Sanford *et al.* It is clear, therefore, that the tendency to cell fusion is not a result of animal passage.

These observations suggest that hybridization *in vitro* of other types of somatic cells should be equally possible. If this hope is justified, hybridization may become a useful tool for the investigation of a number of problems of somatic cell genetics, of oncology and of virology[6]. Examples of problems which are common to these fields and which hybridization *in vitro* could help resolving are: the genetic mechanism of variations in resistance to drugs and other chemotherapeutic agents observed frequently in tumour cells[7]; and the possible occurrence of zygotic induction[8] in virogenic cell lines.

The practicability of the method will depend to a large extent on the establishment of selective techniques for the detection of hybrid cells. Its applicability to the study of the mechanisms of cellular differentiation will require further the establishment of techniques for overcoming barriers of tissue specificity.

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FORTHCOMING EVENTS

*(Meetings marked with an asterisk * are open to the public)*

Monday, May 15

UNIVERSITY OF LONDON (at the Wright–Fleming Institute of Microbiology, St. Mary's Hospital, Paddington, London, W.2), at 5 p.m.—Dr. P. A. Gorer: "Genetics and the Immunology of Cancer".*

BRITISH SOCIETY FOR THE PHILOSOPHY OF SCIENCE (in the Joint Staff Common Room, University College, Gower Street, London, W.C.1), at 5.30 p.m.—Dr. E. W. Bastin: "Self-organizing Mechanisms as Models for Scientific Theories".

INSTITUTION OF ELECTRICAL ENGINEERS, ELECTRONICS AND COMMUNICATION SECTION (at Savoy Place, London, W.C.2), at 5.30 p.m.—Scientific Papers.

UNIVERSITY OF LONDON (in the Anatomy Theatre, University College, Gower Street, London, W.C.1), at 5.30 p.m.—Prof. M. S. Bartlett: "Probability, Statistics and Time" (Inaugural Lecture).*

UNIVERSITY OF LONDON (at King's College, Strand, London, W.C.2), at 5.30 p.m.—Prof. A. Engström (Karolinska Institute, Stockholm): "Micro-X-ray Analysis Applied to Biology and Medicine".* (First of three special university lectures in Biophysics. Further lectures on May 16 and 17.)

ROYAL SOCIETY OF ARTS (at John Adam Street, Adelphi, London, W.C.2), at 6 p.m.—Mr. Lionel Salter: "The Presentation of Music". (Last of three Cantor Lectures on "The Presentation of Science and the Arts on Television".)

INSTITUTION OF ELECTRICAL ENGINEERS, LONDON GRADUATE AND STUDENT SECTION (at Savoy Place, London, W.C.2), at 6.30 p.m.—Annual General Meeting, followed by Dr. P. J. Bowen: "The Experimental Investigation of Space".

Tuesday, May 16

UNIVERSITY OF LONDON (at Birkbeck College, Malet Street, London, W.C.1), at 5.30 p.m.—Prof. G. Martin (University of Bonn): "The Metaphysical Problem of Plato's Theory of Ideas".* (Further lecture on May 18.)

LIBRARIANSHIP STUDENTS ASSOCIATION, NORTH-WESTERN POLYTECHNIC (at the Library Association, Chaucer House, Malet Place, London, W.C.1), at 6.30 p.m.—Prof. Hyman Levy: "The Popularization of Science—an Analysis".

Tuesday, May 16—Thursday, May 18

INSTITUTE OF PHYSICS AND THE PHYSICAL SOCIETY, COLOUR GROUP, and the INTER-SOCIETY COLOR COUNCIL OF THE U.S.A. (at the Royal Institution, Albemarle Street, London, W.1, and Imperial College, London, S.W.7)—Maxwell Colour Centenary.

Wednesday, May 17

INSTITUTE OF PHYSICS AND THE PHYSICAL SOCIETY, LOW TEMPERATURE GROUP (at 47 Belgrave Square, London, S.W.1), at 4 p.m.—Mr. M. Zvegintzov: "Very Low Temperature Engineering".

ROYAL STATISTICAL SOCIETY (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 4.45 p.m.—Dr. P. V. Sukhatme: "The World's Hunger and Future Needs in Food Supplies".

GEOLOGICAL SOCIETY OF LONDON (at Burlington House, Piccadilly, London, W.1), at 5 p.m.—Dr. P. G. Cooray: "The Mode of Occurrence and Geological Significance of the Charnockites of Ceylon". (To be read by Prof. J. Sutton.)

ROYAL METEOROLOGICAL SOCIETY (at 49 Cromwell Road, London, S.W.7), at 5 p.m.—Scientific Papers.

ROYAL SOCIETY OF MEDICINE, COMPARATIVE MEDICINE SECTION (at 1 Wimpole Street, London, W.1), at 5 p.m.—Dr. J. L. Stafford: "Iron Metabolism in Man and Animals"; Dr. D. J. Perkins: "Transferrins and Iron Transport".

UNIVERSITY OF LONDON (at King's College, Strand, London, W.C.2), at 5.30 p.m.—Prof. A. Engström: "Dose–Damage Relationship for Internal Contamination with Radioisotopes".*

BRITISH INSTITUTION OF RADIO ENGINEERS, TELEVISION GROUP (at the London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 6 p.m.—Discussion on "Television Wire Distribution Systems".

Thursday, May 18

ROYAL SOCIETY (at Burlington House, Piccadilly, London, W.1), at 4.30 p.m.—Prof. B. Katz, F.R.S.: "The Transmission of Impulses from Nerve to Muscle, and the Subcellular Unit of Synaptic Action" (The Croonian Lecture).

UNIVERSITY OF LONDON (at the School of Pharmacy, 29–39 Brunswick Square, London, W.C.1), at 5.30 p.m.—Prof. D. Mazia (University of California): "The Analysis of Cell Reproduction".*

ROYAL AERONAUTICAL SOCIETY, ASTRONAUTICS AND GUIDED FLIGHT SECTION (at 4 Hamilton Place, London, W.1), at 6 p.m.—Mr. R. Smelt: "The Agena Satellite and the Discoverer Programme".

INSTITUTION OF ELECTRICAL ENGINEERS (at Savoy Place, London, W.C.2), at 5.30 p.m.—Annual General Meeting. 6.30 p.m.—Mr. J. A. Ratcliffe, F.R.S.: "Experimental Investigation of Space".

ROYAL SOCIETY OF TROPICAL MEDICINE AND HYGIENE (at Manson House, 26 Portland Place, London, W.1), at 7.30 p.m.—Dr. W. E. Ormerod: "The Study of Volutin Granules in Trypanosomes".

Friday, May 19

UNIVERSITY OF LONDON (in the Physiology Theatre, Guy's Hospital Medical School, St. Thomas's Street, London, S.E.1), at 5 p.m.—Prof. W. M. Smart: "Some Aspects of Cosmology" (Fison Memorial Lecture).

INSTITUTION OF ELECTRONICS (in the Large Lecture Theatre, London School of Hygiene and Tropical Medicine, Keppel Street, Gower Street, London, W.C.1), at 7 p.m.—Mr. C. F. Whitbread: "Aerial Techniques".

APPOINTMENTS VACANT

APPLICATIONS are invited for the following appointments on or before the dates mentioned:

GRADUATE RESEARCH ASSISTANTS (2) (with an honours degree in botany, biochemistry or chemistry) IN THE DEPARTMENT OF BOTANY, to participate in research on the mechanism and biochemistry of photosynthesis—The Registrar, Queen Mary College (University of London), Mile End Road, London, E.1 (May 20).

LECTURER (with qualifications in biochemistry, and research and teaching experience) IN THE DEPARTMENT OF CHEMISTRY—The Clerk to the Governors, Chelsea College of Science and Technology, Manresa Road, London, S.W.3 (May 22).

LECTURER IN PURE MATHEMATICS—The Registrar and Secretary, King's College (University of Durham), Newcastle upon Tyne 1 (May 24).

STAFF FELLOW IN THE DEPARTMENT OF PHYSICS, to contribute substantially to the research programme in low temperature physics being initiated in the Department, or to one of the existing programmes in nuclear physics and the physics of solids—The Assistant Registrar (Science), The University, Edgbaston, Birmingham 15 (May 24).

LECTURER or ASSISTANT LECTURER IN BACTERIOLOGY—The Registrar, The University, Leeds 2 (May 27).

LECTURER (preferably with a special interest in geomorphology) IN GEOGRAPHY—The Registrar (Room 22, O.R.B.), The University, Reading (May 27).

LECTURER (with a good honours degree in metallurgy, and preferably some experience in research and industry) IN METALLURGY—The Registrar, The University, Nottingham (May 27).

RESEARCH ASSISTANT (graduate or prospective graduate, with a special interest in the field of growth hormones) IN THE DEPARTMENT OF BOTANY, to undertake work on the physiology of ripening and senescence in annual plants—The Registrar, University College of Wales, Aberystwyth (May 27).

ASSISTANT LECTURER IN BOTANY—The Secretary, Birkbeck College (University of London), Malet Street, London, W.C.1 (May 29).

LECTURER IN PHYSICS—The Registrar, The University, Hull (May 31).

OFFICIAL TUTORIAL FELLOW IN ENGINEERING—The Secretary, St. Catherine's College, 42 Banbury Road, Oxford (May 31).

ASSISTANT LECTURER IN CHEMISTRY tenable in the Department of Physiology—The Secretary, Royal Veterinary College (University of London), Royal College Street, London, N.W.1 (June 3).

ANALYST (DAIRY CHEMIST) (with a degree in science or agricultural science, or equivalent qualifications with emphasis on chemistry) IN THE DEPARTMENT OF AGRICULTURE, Hawkesbury Agricultural College, Richmond, New South Wales, Australia, for investigational work associated with dairy factory manufacturing problems—New South Wales Agent-General's Office, 56 Strand, London, W.C.2 (June 5).

LECTURER or ASSISTANT LECTURER IN APPLIED MATHEMATICS; and a LECTURER or ASSISTANT LECTURER IN PURE or APPLIED MATHEMATICS—The Registrar, The University, Hull (June 5).

LECTURERS or ASSISTANT LECTURERS IN CHEMISTRY AND MATHEMATICS at the Royal University of Malta—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (June 10).

SENIOR LECTURER or LECTURER (preferably with a knowledge of tropical soils) IN SOIL SCIENCE IN THE DEPARTMENT OF AGRICULTURE, University College, Ibadan, Nigeria—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (June 10).

LECTURER IN PSYCHOLOGY at Victoria University of Wellington, New Zealand—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (New Zealand and London, June 15).

LECTURER or ASSISTANT LECTURER (preferably with special qualifications and interests in protozoology, or in the ecology of terrestrial animals, or with special qualifications and interests in cytology and genetics) IN ZOOLOGY at the University College of Ghana—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (June 15).

LECTURER IN VETERINARY PHYSIOLOGY at the University of Sydney, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, June 16).

LECTURER IN PHILOSOPHY in the School of General Studies, Australian National University, Canberra—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, June 17).

SENIOR LECTURER IN AGRICULTURAL ENGINEERING at the University College of Ghana—The Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1 (June 20).

CHAIR OF PHYSICS (ELECTRONIC COMPUTING) in the University of Sydney, Australia—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, June 30).

SENIOR LECTURER IN PHYSICS at the Royal Australian Air Force Academy (University of Melbourne)—The Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1 (Australia and London, June 30).

ASSISTANT LECTURER (Grade B) IN MATHEMATICS—The Principal, Medway College of Technology, Horsted, Maidstone Road, Chatham, Kent.

CHEMICAL RESEARCH TECHNICIAN/ASSISTANT, to work on steroids with a team investigating endocrinology in relation to human biology and genetics—Dr. J. M. Tanner, Institute of Child Health (University of London), The Hospital for Sick Children, London, W.C.1.

ENTOMOLOGIST (with a university degree in science or agricultural science with specialization in entomology, some qualifications in plant pathology, and preferably experience in either agriculture or pest control) in the British Solomon Islands Protectorate, for entomological work in crop protection with special reference to control of amblypelta and brontispa causing damage to coconut palms; work will also cover insect pests of rice, stored copra, village food crops and cocoa, a new crop in the Protectorate—The Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD.63/45/04.

RESEARCH ASSISTANT to help with current research on an immunological project concerned with rheumatoid arthritis—The Pathologist, Stoke Mandeville Hospital, Aylesbury, Bucks.

RESEARCH ASSISTANTS (2) (with a first- or second-class honours degree in geology) IN THE DEPARTMENT OF GEOLOGY—The Clerk to the Governors, Chelsea College of Science and Technology, Manresa Road, London, S.W.3.

SENIOR TECHNICIAN (with experience in general laboratory work, photomicrography and histological techniques) IN THE DEPARTMENT OF ZOOLOGY—The Bursar, The University, Sheffield.

REPORTS and other PUBLICATIONS

(not included in the monthly Books Supplement)

Great Britain and Ireland

The Edinburgh School of Agriculture. (Brochure to commemorate the official opening of the school by H.R.H. The Prince Philip, Duke of Edinburgh, K.G., K.T., LL.D., F.R.S.) Pp. 48 + 8 plates. (Edinburgh: Edinburgh School of Agriculture, 1960.) [172

Parliamentary and Scientific Committee. Annual Report, 1960. Pp. 22. (London: Parliamentary and Scientific Committee, 1961.) [172

University of Leeds. Annual Report of the Librarian, Session 1959–60. Pp. 12. (Leeds: The Brotherton Library, The University, 1960.) [172

Transactions of the Royal Society of Edinburgh. Vol. 64, No. 13 (1959–60): On the Structure of *Deltasperma fouldenense* gen. et sp. nov., and *Cataplosperma berwickense* gen. et sp. nov., Petrified Seeds from the Calciferous Sandstone Series of Berwickshire. By Albert G. Long. Pp. 281–295 + 4 plates. (Edinburgh: Royal Society of Edinburgh, 1961.) 8s. 6d. [172

National Museums of Southern Rhodesia. Ichthyology: The Fish of the Middle Zambesi. By P. B. N. Jackson. (Kariba Studies.) Pp. ii + 36. (Manchester: Manchester University Press; New York: Humanities Press, Inc., 1961.) 6s. net. [172

British Museum (Natural History). Ruwenzori Expedition, 1952. Vol. 1, No. 7: Noctuidae. By D. S. Fletcher. Pp. 177–323 (325 figures). (London: British Museum (Natural History), 1961.) 40s. net. [172

Dunsink Observatory Publications, Vol. 1, No. 1: Cape Lyot Heliograph Results. 1: Light-Curves of 30 Solar Flares in Relation to Sudden Ionospheric Disturbances. By M. A. Ellison, Susan M. P. McKenna and J. H. Reid. Pp. 36 + 2 plates. (Dublin: Dublin Institute for Advanced Studies, 1961.) [172

Planning, Vol. 27, No. 449 (20 February, 1961): Systems of Measurement. Pp. 33–56. (London: Political and Economic Planning, 1961.) 3s. [202

Other Countries

Chicago Natural History Museum. Fieldiana: Botany. Vol. 29, No. 5: A Revision of Cedrela (Meliaceae). By C. Earle Smith, Jr. Pp. ii + 295–341 + plates 7–14. 1.75 dollars. Fieldiana: Geology, Vol. 14, No. 3: *Aretocyclus* and Some other Chadronian Vertebrate Microfossils from Nebraska. By William D. Turnbull and Charles A. Reed. Pp. 41–57. 70 cents. Fieldiana: Zoology, Vol. 39, No. 42: *Sanala pauciramus*, a New Gonostomatid Fish from the Western Atlantic. By Marion Grey. Pp. 465–471. 20 cents. Vol. 39, No. 43: A Review of the Oriental Toads of the Genus *Ansonia* Stoliczka. By Robert F. Inger. Pp. 473–503. 95 cents. Vol. 42, No. 8: Philippine Zoological Expedition, 1946–1947. On Some Parasitic Laelaptoid Mites (Acarina) of the Philippines. By Mercedes D. Delfinado. Pp. 93–114. 75 cents. (Chicago: Chicago Natural History Museum, 1960.) [172

United Nations Educational, Scientific and Cultural Organization. Arid Zone Research. Plant-Water Relationships in Arid and Semi-Arid Conditions: Reviews of Research. Pp. 225. (Paris: Unesco; London: H.M. Stationery Office, 1960.) 21 NF.; 30s. 6d.; 6 dollars. [92

Istituto di Microbiologia, Università, Milano. The Culture Collection of the Progetto Sistematica Actinomiceto, 1961. Pp. 4 + 11. (Milano: Istituto di Microbiologia, Università, 1961.) [102

Meddelanden fran Oceanografiska Institutet I Göteborg, No. 27: Transparency Measurements in the Baltic Sea. By Masaaki Fukuda. Pp. 17. No. 28: Gesteinsfragmente im Roten ton des Atlantischen Ozeans (N 29° 21', W 58° 09', Tiefe 5 450 M). Von Otto Mellis. Pp. 18. (Göteborg: Elanders Boktryckeri Aktiebolag, 1960.) [102

National Research Development Corporation of India. Sixth Annual Report and Statement of Accounts for the period ending 31st March, 1960. Pp. 25. (New Delhi: National Research Development Corporation of India, 1960.) [102

National Science Foundation, Washington. Tenth Annual Report for the Fiscal Year ended June 30, 1960. Pp. xiii + 310. (Washington, D.C.: Government Printing Office, 1961.) 1 dollar. [102

United States Department of Health, Education and Welfare, Public Health Service Publication No. 757: Division of General Medical Sciences, National Institutes of Health. Pp. viii + 23. Descriptive brochure. (Washington, D.C.: Government Printing Office, 1961.) 15 cents. [102

Annals of the New York Academy of Sciences, Volume 89, Article 4: Thermodynamics and Mechanics of Polymer Systems. By F. R. Eirich and 9 other authors. Pp. 573–774. (New York: New York Academy of Sciences, 1961.) [102

City of Durban. Annual Report of the Durban Museum and Art Gallery for the Municipal Year 1959–1960. Pp. 20. Durban Museum Novitates, Vol. 6, Part 3 (15th October, 1960): Relationships within the *Camaroptera fasciolata*—Stierlingi—Simplex Complex of Warblers, by Michael P. Stuart Irwin. Pp. 47–60. 3s. net. Vol. 6, Part 4 (21st October, 1960): A New Species of Canary from Southern Portuguese East Africa. By P. A. Clancey and W. J. Lawson. Pp. 61–64. 1s. net. Vol. 6, Part 5 (20th December, 1960): Notes on Some Central and East African Bats. 1: The Occurrence of Bocage's Fruit Bat *Rousettus* (*Lissonycteris*) *Angolensis* Bocage, 1898, in Southern Rhodesia. By David L. Harrison. Pp. 65–78. 2s. 6d. net. (Durban: Durban Museum and Art Gallery, 1960.) [102

Archives de l'Institut de Paléontologie Humaine (Fondation Albert Ier, Prince de Monaco). Mémoire No. 31: La Grande Roche de Naquane. Par E. Anati. Pp. 189 (52 planches). (Paris: Masson et Cie., 1961.) [102

United Nations Educational, Scientific and Cultural Organization. Centro de Co-operacion Cientifica para America Latina. Scientific Institutions and Scientists in Latin-America. Argentina (Scientificos) (IV Volumen), Fasciculo V. Pp. i + 707–767. (Montevideo: UNESCO; Centro de Co-operacion Cientifica para America Latina, 1960.) [142

National Institute of Sciences of India. Chemistry of Bi- and Tripositive Silver. By P. Ray and D. Sen. (N.I.S.I. Monographs, Silver Jubilee Issue.) Pp. iv + 33. (New Delhi: National Institute of Sciences of India, 1960.) Rs. 4.00. [172

Publications de l'Institut National pour l'Étude Agronomique du Congo. Carte des Sols et de la Vegetation du Congo et du Ruanda-Urundi, 11: Ubangi. A: Sols. B: Vegetation. C: Utilisation des Sols. D: Geomorphologie. Notice Explicative de la Carte des Sols et de la Vegetation. Par P. Jongen, M. Van Oosten, C. Evrard et J. M. Berce. Pp. 82. 17: Region de Yanonge-Yatolema. A: Sols. Notice Explicative de la Carte des Sols. Par A. Van Wambeke. Pp. 28. (Bruxelles: Institut National pour l'Étude Agronomique du Congo Belge, 1960.) [172

International Hydrographic Bureau, Monaco. Supplementary Paper I to Special Publication 39: Radio Aids to Maritime Navigation and Hydrography. Chapter V: Tellurometer. Micro-Dist. Pp. iv + 39. Supplementary Paper 2 to Special Publication 39: Radio Aids to Maritime Navigation and Hydrography. Chapter III: Raydist. Hydrodist. M.P.F.S., Derveaux. Pp. ii + 6s. (Monaco: International Hydrographic Bureau, 1960.) [172

Somaliland Protectorate: Geological Survey. Report No. 4: Report on the Geology of the Berbera-Sheikh Area, Berbera and Burao Districts (Quarter Degree Sheets Nos. 24 and 36). D.O.S. 1: 125,000). By John A. Hunt. Pp. vii + 27. (Hargeisa: Stationery Office; London: Crown Agents for Oversea Governments and Administrations, 1960.) 8s. 20. [172

Università di Genova, Istituto Geofisico e Geodetico con annesso Osservatori Geofisici di Genova. Relazione sull' Attivita Svolta nel 1960. Elenco delle Pubblicazioni 1947–1960. Pp. 13. (Genova: Università di Genova, Istituto Geofisico e Geodetico, 1960.) [202

Norsk Polarinstitutt. Meddelelser, Nr. 83: Reinen på Svalbard. Av Odd Lønø. Pp. 31. Kr. 2.00. Nr. 85: Some Dinoflagellates and Hystrichosphaerida from the Lower Tertiary of Spitsbergen. By Svein Manum. Pp. 9 + 1 plate. Kr. 2.00. Skrifter, Nr. 119: Ralus Slopes and Mountain Walls at Tempelfjorden, Spitsbergen—a Geomorphological Study of the Denudation of Slopes in an Arctic Locality. By Anders Rapp. Pp. vii + 96 + 20 plates. Kr. 25.00. (Oslo: Norsk Polarinstitutt, 1960. Distributed by the Oslo University Press.) [202

Académie Royale de Belgique. Classe des Sciences. Mémoires, Tome 32, Fascicule 4: Réactions Directes et Modèles Nucléaires. Par Marcel Demeur. Pp. 40. (Bruxelles: Académie Royale de Belgique, 1960.) 40 francs. [202

National Academy of Sciences—National Research Council. Nuclear Science Series. NAS–NS 3015: The Radiochemistry of Zinc. By Harry G. Hicks. Pp. vi + 58. 75 cents. NAS–NC 3016: The Radiochemistry of Protactinium. By H. W. Kirby. Pp. vi + 80. 1 dollar. NAS–NS 3017: The Radiochemistry of Iron. By J. M. Nielson. Pp. v + 42. 50 cents. (Washington, D.C.: Office of Technical Services, Department of Commerce, 1960.) [202

Cawthron Institute, Nelson, New Zealand. Radioactivity in the Service of Man. By Dr. J. F. Duncan. (Thomas Cawthron Memorial Lecture, No. 34, 1959.) Pp. 43. Thomas Cawthron Memorial Lectures, No. 32: New Mineral Discoveries in the Nelson Province. By Dr. Gordon J. Williams. Pp. 29. No. 33: The New Zealand Antarctic Expedition. By Sir Edmund Hillary. Pp. 12 + 4 plates. (Nelson: Cawthron Institute, 1957, 1958 and 1960.) [202

Kybernetik, Band 1, Heft 1 (January, 1961). (A journal dealing with the Transmission and Processing of Information as well as with Control Processes in both Organisms and Automata.) Pp. 56. (Berlin: Springer-Verlag, 1961.) 12.80 D.M. [202

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APPOINTMENTS VACANT

NEW ZEALAND

EDUCATION DEPARTMENT

Applications are invited for the following positions at the New Zealand School of Pharmacy:

SENIOR TUTOR OR TUTOR IN PHARMACEUTICS

Applicants should possess a degree or registrable qualification in pharmacy and have had teaching experience. A postgraduate qualification and/or research experience would be an additional advantage. The appointee will be in charge of all the teaching in pharmaceutics and will be particularly responsible for the teaching of pharmaceutical microbiology.

TUTOR OR ASSISTANT TUTOR IN PHARMACOGNOSY

Applicants should possess a degree or registrable qualification in pharmacy. A postgraduate qualification, teaching or research experience would be an additional advantage. The appointee will be responsible for all the teaching in pharmacognosy and should also be prepared to assist with the teaching of pharmaceutics.

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Incidental Expenses: Up to £35 for a single person and £100 for a married man can be claimed to cover the cost of taking personal effects to New Zealand.

Inquiries, quoting reference number B11/8/9, should be addressed to the High Commissioner for New Zealand, 415 Strand, London, W.C.2, who will supply further details and application forms.

Completed applications to be lodged with this office not later than May 29, 1961.

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UNIVERSITY OF LEEDS

DEPARTMENT OF BACTERIOLOGY

Applications are invited for appointment as Lecturer or Assistant Lecturer in Bacteriology. Salary on the clinical scale for a Lecturer is £1,350 by £100 to £1,950 (efficiency bar) by £100 to £2,350, or on the scale £1,050 by £50 to £1,400 by £75 to £1,850 (efficiency bar £1,550) if non-medically qualified, and for an Assistant Lecturer on the clinical scale £1,050 by £100 to £1,250, or on the scale £800 by £50 to £950 if non-medically qualified.

Applications (three copies), stating date of birth, qualifications and experience, together with the names of three referees, should reach the Registrar, the University, Leeds, 2 (from whom further particulars may be obtained) not later than May 27, 1961.

UNIVERSITY OF LEEDS

DEPARTMENT OF PHYSIOLOGY

Applications are invited for appointment as Demonstrator (Assistant Lecturer) in Physiology at a salary on the scale £1,050 by £50 to £1,150 a year if medically qualified, or within the range £800 to £950 a year if not medically qualified.

Applications (three copies), stating date of birth, qualifications and experience, together with the names of three referees, should reach the Registrar, the University, Leeds, 2 (from whom further particulars may be obtained) not later than May 29, 1961.

MENTAL HEALTH RESEARCH FUND FELLOWSHIPS

Applications are invited for Mental Health Research Fund Fellowships from suitably qualified persons wishing to pursue full-time research work bearing on problems of mental health, whether in clinical psychiatry or one of its supporting sciences. Both Junior and Senior Fellowships are offered in the salary ranges £700 to £1,805 and £1,800 to £2,500 plus superannuation. The appointments will be for up to three years in the first instance.

Application forms, which must be returned before October 1, 1961, may be obtained, together with further information, from the Secretary, Research Committee, Mental Health Research Fund, 39 Queen Anne Street, London, W.1 (Tel.: WELbeck 1272).

UNIVERSITY COLLEGE OF SOUTH WALES AND MONMOUTHSHIRE

Applications are invited for the post of Lecturer or Assistant Lecturer in the Department of Physiology from candidates (medically qualified or otherwise).

Further particulars may be obtained from the Registrar, University College, Cathays Park, Cardiff, to whom six copies of applications (including the names of three referees) are to be sent not later than Monday, June 19, 1961.

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Applications, stating age, qualifications, together with the names of two referees, should be sent to the Group Secretary, General Hospital, Nottingham.

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Transport:	Reasonable amount allowed for transport of appointee and dependent family to Western Australia provided bond entered into to serve State for period of three years.
Conditions of Service:	Superannuation benefits, three months' long service leave after seven years' continuous service, cumulative sick leave entitlements and other general conditions applicable to permanent public servants.

Applications, stating age, qualifications and experience, should be addressed by June 30, 1961, to:

Public Service Commissioner,
184 St. George's Terrace,
PERTH, Western Australia.

COUNTY OF ESSEX
EDUCATION COMMITTEE
RUSH GREEN COLLEGE OF FURTHER EDUCATION
DAGENHAM ROAD, RUSH GREEN

Required August 1, 1961, at this new college: (i) Senior Laboratory/Workshop Technician. Salary £685 by £25 (3) to £760 per annum, plus London allowance not exceeding £40 per annum. (ii) Laboratory/Workshop Technicians (three posts). Salary within range £490 to £685 per annum, depending on qualifications and experience, plus London allowance not exceeding £40 per anum. Permanent, superannuable posts. Forty-two hour week. For the senior post applicants will be expected to have good laboratory or workshop experience, and possession of an appropriate City and Guilds or other qualification will be an advantage. Applicants for the other three posts should preferably have had industrial, technical or workshop experience. Applications would be considered from candidates under 21, to whom a special salary scale applies (£260 to £420 depending on age). Technicians are encouraged to improve their qualifications after appointment.

Apply by letter to the Chief Education Officer (Ref. F.7.k.), County Offices, Chelmsford. Closing date May 22, 1961.

AUSTRALIAN NATIONAL UNIVERSITY
CANBERRA
JOHN CURTIN SCHOOL OF MEDICAL RESEARCH
DEPARTMENT OF MEDICAL CHEMISTRY

Applications are invited from organic chemists for a Research Fellowship in the Department of Medical Chemistry. The successful candidate will work with Professor Adrien Albert or Dr. D. J. Brown on problems in the pyrimidines, pteridines or other hetero-aromatic series. The salary range for Research Fellows is £A.1,625 to £A.2,450 per annum, and appointment is made for three years, with possible extension to the maximum of five years. Reasonable travel expenses will be paid and assistance with housing provided. Superannuation is on the F.S.S.U. pattern.

Further particulars should be obtained from the Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1. Applications close, in Australia and London, on June 30, 1961.

ICI

IMPERIAL CHEMICAL INDUSTRIES LIMITED

Physiologists and Pharmacologists

Opportunities are offered for young physiologists and pharmacologists to take part in important investigations concerning drugs affecting the circulatory, reproductive and central nervous systems. Candidates should be of sound academic qualification with aptitudes and training for research and with enthusiasm and originality of thought.

Appointments will be offered in modern, well-equipped laboratories situated in rural Cheshire seventeen miles south of Manchester and will provide facilities for close collaboration with chemical, biochemical, pathological and microbiological colleagues. Presentation of all worthwhile discoveries at academic meetings and their publication in scientific periodicals is encouraged.

Inquiries, with brief details of qualifications and experience, should be sent to

The Staff Manager,
I.C.I. Limited, Pharmaceuticals Division,
Fulshaw Hall, Wilmslow, Cheshire.

CHEMISTS
PHYSICISTS
TECHNICIANS

3M COMPANY

MINNESOTA MINING AND MANUFACTURING COMPANY

require experienced organic, physical and inorganic chemists (including emulsion chemists), physicists and technicians for research and development work in England or U.S.A.

The research will be concerned initially with the development of new photographic materials and a number of **SENIOR POSITIONS** are open.

Applications, giving details of age, qualifications and experience, which will be treated in strict confidence, should be made in writing, to:

Dr. J. D. Kendall, Director, Minnesota 3M Research Limited, 3M House, Wigmore Street, London, W.1

UNIVERSITY COLLEGE OF NORTH STAFFORDSHIRE
KEELE, STAFFORDSHIRE

Applications are invited for a postgraduate Research Studentship for work in Biogeography: Geobotany in Northern Australia, tenable at the University College of North Staffordshire for two years from October 1, 1961. The value of the studentship will be between £600 and £800 per annum depending on qualifications, plus travelling expenses to and from Australia and field expenses in Australia. Candidates should preferably possess qualifications in geography and botany or botany and geology, but those offering one of the first two alone will also be considered.

The successful applicant will work under the supervision of Dr. M. M. Cole, to whom applications should be sent before May 20, 1961.

UNIVERSITY OF BRITISH COLUMBIA
BIOPHYSICIST
METEOROLOGIST

The Physics Department invites inquiries for vacancies at the Assistant Professor rank in (a) Biophysics and (b) Meteorology. In each case the position involves both teaching and research and the potential in these areas will be given greater weight than experience. The appointee will be given an opportunity to initiate his own programme of course work and research. The minimum salary for an Assistant Professor is $7,000. The appointment is required for the Session commencing in September.

Those interested should address their inquiries to: G. M. Shrum, Physics Department, University of British Columbia, Vancouver 8, B.C.

CHEMISTS AND BIOCHEMISTS

The basic research group of the Unilever Research Laboratory, Colworth House, has vacancies for PHYSICAL and ORGANIC CHEMISTS and BIOCHEMISTS to work on interesting and fundamental problems. Colworth House is a large and rapidly expanding research laboratory (present total staff 525) and is situated in a 1,200-acre country estate 10 miles north of Bedford. The basic research group is mainly engaged in long-term research to supply the fundamental information necessary for the expansion of the Unilever food and animal feedingstuffs interests during the next 5 to 15 years.

Problems on which we are working are: the investigation of enzyme systems; plant biochemistry; the isolation, characterization and synthesis of natural flavouring material; the biochemistry of meat; biochemical investigation of digestion and nutrition in collaboration with a newly-established physiology unit; and the physical properties of plant and animal proteins.

Applications from newly qualified scientists or those with up to 15 years academic or industrial research experience are invited. The salary for a newly qualified graduate with a first or second-class honours degree is £800 per annum; for a graduate with three years research experience £1,025 per annum; greater experience will command a correspondingly higher salary.

Please apply to the Staff Officer (FRR. 116),
UNILEVER RESEARCH LABORATORY,
COLWORTH HOUSE, SHARNBROOK, BEDFORD.

AUSTRALIAN NATIONAL UNIVERSITY

CANBERRA

INSTITUTE OF ADVANCED STUDIES
DEPARTMENT OF ASTRONOMY

The University proposes to make three appointments in the Department of Astronomy (Head of Department: Professor B. J. Bok, Mount Stromlo Observatory). Each appointment will be made at one of several levels between those of Senior Fellow and Research Fellow, dependent on the qualifications and experience of the appointee. The salary ranges are: Senior Fellow, £A.3,000 to £A.3,550; Fellow, £A.2,120 to £A.3,000; Senior Research Fellow, £A.2,620 to £A.3,160; Research Fellow, £A.1,625 to £A.2,450. Superannuation is on the F.S.S.U. pattern. Reasonable travel expenses are paid and assistance with housing is provided.

A summary of conditions of appointment and descriptive papers setting out the research programme and facilities of Mount Stromlo Observatory and the Department of Astronomy, A.N.U., are available from the Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1. Applications will be considered immediately after June 1 and October 1, 1961.

CHELSEA COLLEGE OF SCIENCE AND TECHNOLOGY

MANRESA ROAD, LONDON, S.W.3
DEPARTMENT OF CHEMISTRY

Applications are invited for the post of Lecturer from candidates possessing (i) qualifications in Biochemistry, and (ii) research and teaching experience. Teaching duties for the present will be in connection with the College Diploma in Biochemistry. Salary scale: £1,370 by £35 to £1,550, plus London allowance of £38 or £51, according to age.

Particulars and forms of application from the Clerk to the Governors, to whom applications should be submitted not later than May 22, 1961.

LEVINGTON RESEARCH STATION

DEPARTMENT OF SOIL SCIENCE

FISONS FERTILIZERS LIMITED invite applications for the interesting and responsible appointment of Agricultural Adviser in the provinces on the staff of Levington Research Station.

Applicants should be graduates in Agriculture and be well informed on matters of specialist agricultural advice. Experience of field experimentation will be advantageous as the appointment is concerned mainly with this subject.

An Agricultural Adviser resident in a provincial location is provided with a car and has a Crop Recorder attached to him for assisting with field experiments.

This is a senior appointment and the salary offered will depend upon qualifications and experience. There are contributory pension and widow's pension schemes.

Applications, giving qualifications and experience, should be made to the Personnel Manager (LN10), Fisons Fertilizers Limited, Harvest House, Felixstowe.

UNIVERSITY OF OTTAWA

OTTAWA, ONTARIO, CANADA

Assistant Professor in Pharmacology required. Ph.D. or M.B. Salary $6,000 to $8,500, according to qualifications and experience.

Applications to Professor M. F. Murnaghan at the above address.

NEW ZEALAND

DEPARTMENT OF SCIENTIFIC AND INDUSTRIAL RESEARCH
VACANCIES FOR PLANT BREEDERS
CROP RESEARCH DIVISION

The Crop Research Division has vacancies for graduates with appropriate honours degrees for plant breeding work, at Lincoln in the South Island and at Otara in the North Island.

Vacancy No. 1260. Field Crops Section—Lincoln. To participate in a programme of plant improvement of forage crops, including breeding for resistance to fungus, virus and aphid attack in Brassicas and for increased winter growth in lucerne. In the Brassicas interspecific hybridization is being used to transfer characters of economic importance, and an active research programme on economic and intrinsic interest has been developed on the genetics, cytology and fertility of these hybrids. The officer appointed would have the opportunity of carrying out research in genetics, evolution or cytology, not necessarily in the fields of his plant breeding work. The Division is situated 13 miles from Christchurch and half a mile from Canterbury Agricultural College.

Vacancy No. 1261. Vegetable Section—Otara. To work on the improvement of vegetable varieties for processing (freezing and canning), in particular, peas, beans, tomatoes and asparagus. The officer appointed will be required to develop a programme of plant breeding on the basis of a survey at present being conducted. Training and/or experience in post-harvest physiology of vegetable crops would be an advantage since selection for quality and against deterioration in the field-to-factory and processing phases may be of major significance. There will be considerable scope for original research in this field. The appointee will be one of a team which deals also with general improvement of market varieties, disease resistance in field and glasshouse vegetables, seed production and related problems, research in the ethnobotanical field and cytology and evolution in some Pacific domesticated plants. The Vegetable Research Station is situated at Otara, near Auckland.

Salary: The salary offered for these positions is up to £1,700 a year, according to qualifications and experience and further advancement is on scientific merit. An outstanding officer can ultimately advance to £2,300 a year without administrative responsibility.

Fares payable: United Kingdom. Up to £165 each for steamer fares for appointee and wife and pro rata for dependent children plus fares to port of embarkation and from port of arrival to destination in New Zealand. South Africa. Up to £210 each for steamer fares for appointee, etc., as above for United Kingdom. Other countries. Minimum first-class rail and/or steamer fares by shortest and most direct route to New Zealand. U.K. Incidental Expenses. Up to £35 for a single person and £100 for a married man can be claimed to cover the cost of taking personal effects to New Zealand.

Application forms and further details are available from the High Commissioner for New Zealand, 415 Strand, London, W.C.2, with whom applications will close on June 17, 1961. Please quote reference B/13/18/2, when inquiring.

UNIVERSITY OF ABERDEEN

RESEARCH FELLOWSHIP

Applications invited for a Postdoctoral Fellowship (Senior or Junior) for research on the Immunology of the Pituitary Gonadotrophic Hormones to be carried out in the Agricultural Biochemistry Division of the Department of Biological Chemistry. Previous experience of immunological methods desirable but not essential. The appointment would be for one year in the first instance. The value of this Fellowship will be within the range £800 to £1,050 per annum with F.S.S.U.

Applications should be sent to the Secretary, the University, Aberdeen, from whom further particulars may be obtained.

RED CROSS BLOOD TRANSFUSION SERVICE

QUEENSLAND
BIOCHEMIST

Applications are invited for the position of Biochemist to supervise all chemical procedures and to conduct research in fields related to blood transfusion in consultation with the Director. New Transfusion headquarters are in planning stage and the appointee may be required to advise on the design of the biochemical laboratories. Salary within the limits £A.2,500 to £A.3,500 per annum (actual), to be determined in relation to qualifications and experience. Superannuation is available.

Full details may be obtained from the Director, 409 Adelaide Street, Brisbane.

NEW ZEALAND
DEPARTMENT OF SCIENTIFIC AND INDUSTRIAL RESEARCH
SOIL BUREAU

The Soil Bureau of the Department of Scientific and Industrial Research has vacancies for honours graduates in science or agriculture for the positions of soil chemist, pedologist and mycologist. The duties of these positions are:

Vacancy No. 1189 (B13/18/4). Soil Chemist. Research work in the Physical Chemistry Section of the Bureau. The current work of this section includes clay mineralogy in relation to soil processes and the role of different soil clays in nutrient availability. X-ray diffraction, spectrographic, differential thermal analysis techniques are in current use.

Vacancy No. 1190 (B13/18/5). Pedologist. The mapping and classification of soils, pedological research and the application of these findings to land use in co-operation with the agriculturist, forester, town planner, and engineer.

Vacancy No. 1191 (B/13/18/6). Mycologist. The study of soil fungi and their role in soil formation.

The soil chemist will be stationed in Wellington and the mycologist at Taita, about 15 miles from Wellington. The pedologist will be stationed initially at Wellington but after training will be placed in one of the Bureau's field offices. In 1962 Wellington staff will transfer to a new laboratory being built at Taita.

Salary: The salary offered for these positions is up to £1,500 a year according to qualifications and experience and further advancement is on scientific merit. An outstanding officer can ultimately advance to £2,300 a year without administrative responsibility.

Fares payable: United Kingdom. Up to £165 each for steamer fares for appointee and wife and pro rata for dependent children plus fares to port of embarkation and from port of arrival to destination in New Zealand. South Africa. Up to £210 each for steamer fares for appointee and wife and pro rata for dependent children plus fares to port of embarkation and from port of arrival to destination in New Zealand. Other countries. Minimum first-class rail and/or steamer fares by shortest and most direct route to New Zealand. U.K. Incidental Expenses: Up to £35 for a single person and £100 for a married man can be claimed to cover the cost of taking personal effects to New Zealand.

Applications completed after the closing date from countries outside the United Kingdom will be considered.

Application forms and further details are available from the High Commissioner for New Zealand, 415 Strand, London, W.C.2, with whom applications will close on June 17, 1961. Please quote vacancy and reference number when inquiring.

UNIVERSITY OF ALBERTA
FACULTY OF AGRICULTURE

The University of Alberta invites applications for a position in the Department of Animal Science as Assistant Professor (poultry genetics) at a starting salary of $6,600, with excellent prospects for advancement. Candidates must have a Ph.D. or equivalent in genetics. Preference will be given to applicants with training in biochemical genetics. Duties, to commence September 1, 1961, will include fundamental and applied research on methods of predicting production and reproduction traits in poultry, co-operation in research projects on genetic-nutritional-environmental interactions, teaching and limited extension work.

Forward applications, accompanied by a recent photograph, curriculum vitae, transcript of academic record, and names and addresses of three references to the Head, Department of Animal Science, University of Alberta, Edmonton, Alberta. Closing date: June 30, 1961.

UNIVERSITY OF EDINBURGH
DEPARTMENT OF CHEMISTRY

Applications are invited for the post of Lecturer in the above Department. Candidates should have special interests in physical or inorganic chemistry. Salary scale: £1,050 by £50 to £1,250; bar: £1,300 by £50 to £1,400 by £75 to £1,850 per annum, with placement according to qualifications and experience, and with superannuation benefit and family allowance where applicable. The successful candidate will be expected to take up duty on October 1, 1961.

Further particulars may be obtained from the undersigned, with whom applications (six copies), giving the names of two referees, should be lodged not later than May 31, 1961.

CHARLES H. STEWART,
Secretary to the University

Animal Nutrition Research

Research scientists with postgraduate research experience are invited to apply for the following appointments in the expanding Animal Nutrition Division of the UNILEVER RESEARCH LABORATORY at Colworth in North Bedfordshire.

CATTLE NUTRITION

This section investigates fundamentally the nutritional requirements of calves, growing and fattening cattle and milking cows with a view to developing improved foods and systems of feeding. Excellent facilities for metabolism studies and feeding experiments are backed by the resources of the chemical, bacteriological and statistical sections in the other divisions of the laboratory.

There are two posts concerned with the lactation and growth studies. Applicants should have research experience in ruminant nutrition or one of the cognate sciences and be interested in the nutritional physiology of the ruminant and its bearing on the reactions of the animal under normal farming conditions.

POULTRY NUTRITION

The work of this section embraces fundamental growth studies and the application of these results to the production and marketing of poultry and the development of feedingstuffs. A recently occupied new building provides excellent facilities for research.

Applicants should have at least one or two years postgraduate experience and their training should include biochemistry at graduate or postgraduate level and either a veterinary degree or a keen interest in nutritional problems.

Starting salaries for all these appointments will depend upon experience and qualifications. Keen and able staff can expect generous increments in salary and look forward to a satisfying research career.

Please write for an application form to:
THE STAFF OFFICER (ERR. 119), UNILEVER RESEARCH LABORATORY, COLWORTH HOUSE, SHARNBROOK, BEDFORD

UNIVERSITY OF ABERDEEN
DEPARTMENT OF PHYSIOLOGY

Applications invited for: A Senior Research Fellowship, salary not less than £1,100 per annum, plus F.S.S.U. A Junior Research Fellowship, salary within the range £800 to £950 per annum, plus F.S.S.U. Both appointments, which are financed by the U.S. Public Health Service, are for three years, beginning on September 1 or a date to be arranged. The Fellows will form a team, led by Dr. H. W. Kosterlitz, to investigate the mechanism of the action of morphine and other analgesics on the peripheral autonomic nervous system. One Fellow should have some experience in pharmacological techniques and the other in neurophysiological techniques.

Further details and forms of application may be obtained from the Secretary, Marischal College, Aberdeen, to whom applications (three copies), giving details of qualifications, etc., and the names of two referees, should be submitted by May 27, 1961.

APPLICATIONS ARE INVITED FOR THE post of Curator of the Coryndon Museum, Nairobi, Kenya. The appointment will be for a period of three years, in the first instance, commencing on July 1, 1961, or as soon after as possible. Candidates should, if possible, have experience in Museum administration as well as general scientific knowledge, especially in biological fields. The post is principally administrative, but will not entail protracted or frequent absence from the Museum buildings. Salary will be in the scale of £2,000 by £60 to £2,540, the point of entry being decided on the basis of previous experience and qualifications. An overseas appointee will receive economy class, air passage for himself and wife on taking up the appointment and return passages at the end of three years.—Write in the first instance to: The Secretary, Coryndon Museum, P.O. Box 658, Nairobi, Kenya, East Africa, when further details will be supplied if required.

UNIVERSITY COLLEGE OF WALES
ABERYSTWYTH
DEPARTMENT OF BOTANY

Applications are invited for appointment as Research Assistant to undertake work on "The Physiology of Ripening and Senescence in Annual Plants," under a grant from the Agricultural Research Council. Candidates, who should be graduates or prospective graduates, should have a special interest in the field of growth hormones. The person appointed will be eligible to apply for acceptance to work for a higher degree. Salary: £400 (plus superannuation).

Further particulars from the Registrar, to whom applications must be made by May 27.

NEW ZEALAND
DEPARTMENT OF SCIENTIFIC AND INDUSTRIAL RESEARCH

Applications are invited for the undermentioned vacancy: Vacancy No. 1277—Editor or Assistant Editor, Information Bureau, Head Office, Wellington, Department of Scientific and Industrial Research. Salary: Up to £1,700 per year according to qualifications and experience. Qualifications desired: A degree in science with qualifications in geology or geophysics. An interest in a wide range of scientific subjects is desirable. Duties: Editing (or assisting to edit) the "N.Z. Journal of Geology and Geophysics" with responsibilities for its standard. Passages: Steamer fares of up to £165 each for the appointee and his wife, and pro rata for dependent children will be paid. Incidental expenses: Up to £35 for a single person and £100 for a married man can be claimed to cover the cost of taking personal effects to New Zealand.

Application forms and further details are available from the High Commissioner for New Zealand, 415 Strand, London, W.C.2, with whom applications will close on June 30, 1961. Please quote reference B.13/18/3 when inquiring.

HEAVY ORGANIC CHEMICALS DIVISION

is expanding its

EXPLORATORY RESEARCH GROUP

to study new tonnage chemicals, to develop *new ways* of making them, to establish the *mechanisms* by which they are formed and to work out the *kinetics* of the reactions involved.

Men and women are needed for this

EXPLORATORY RESEARCH TEAM

Organic chemists and physical chemists are needed most, but we should be glad to have a physicist interested in building novel equipment, a first-class mathematician keen on process development techniques, and chemical engineers anxious to work with chemists.

For further information please write to:

Exploratory Research Group Manager
Imperial Chemical Industries Limited,
Heavy Organic Chemicals Division,
Billingham-on-Tees, Co. Durham,

quoting reference MB/ER/5.

NATURAL RUBBER PRODUCERS' RESEARCH ASSOCIATION

PHYSICISTS and CHEMISTS

The Natural Rubber Producers' Research Association require physicists and chemists to undertake research on polymers. The appointments are especially suited to those wishing to make their career in research. Candidates should possess a good honours degree, some research experience would be advantageous but it is not essential. A high level of ability and enthusiasm is sought, and successful applicants will be given every encouragement, including a liberal publication policy, to show initiative and responsibility. Salary initially will be within the range £760 to £1,695 according to age, qualifications and experience, with superannuation under the F.S.S.U. scheme, and an allowance for children. Pleasant working conditions in laboratories located about 20 miles north of London.

Please write to the Director of Research, 48-56 Tewin Road, Welwyn Garden City.

Candidates from overseas should include full details with their application.

UNIVERSITY OF CAMBRIDGE

PROPOSED UNIVERSITY SCIENCE LIBRARY
APPOINTMENT OF ASSISTANT UNDER-LIBRARIAN

Applications are invited from graduates in science or with experience of library administration for appointment to a post of Assistant Under-Librarian. The person appointed will initially work in the University Library; and the duties will be the preparation of a classification scheme for the Science Library, the reclassifying of science material at present in the University Library, and assistance in the planning of the new Science Library building. The present salary scale for Assistant Under-Librarians is £800 to £1,550 a year. A non-pensionable allowance of £50 a year is paid for each dependent child who is under the age of 16 or is in receipt of full-time education. The Council of the Senate may give the person appointed a number of years' seniority for the purpose of determining his stipend. A contribution towards removal expenses may be made.

Further particulars may be obtained from the Librarian, University Library, Cambridge, and applications should be sent to him so as to reach him not later than June 22, 1961. Testimonials are not required but a candidate should give the names of two persons to whom reference may be made.

UNIVERSITY COLLEGE OF SWANSEA

BOTANY DEPARTMENT

Applications are invited for the post of Technical Research Assistant to work with Professor Street on the establishment of free cell cultures from higher plants at a salary of £450 per annum as soon as possible.

Applicants should have some training in chemistry and biology and should send an outline of their training and qualifications, together with the name of a Referee, to the Registrar, from whom further particulars may be obtained.

BOVRIL LIMITED

ASSISTANT CHEMIST
MICROBIOLOGIST
LABORATORY ASSISTANT

There are vacancies for the above posts in the Central Control Laboratories.

The work will cover a wide range of food products: and offers interest, scope and prospects. Previous experience in the food industry desirable.

Five-day week, contributory pension scheme, canteen, social club.

Write, giving full details of age, qualifications and experience, to the Group Chemist, 148-166 Old Street, E.C.1.

UNIVERSITY COLLEGE

IBADAN, NIGERIA

Applications are invited for Lectureship in Department of Zoology. Preference given to applicants with a special interest in either vertebrate zoology or genetics. Appointment to commence October, 1961. Salary scale: £1,200 by £75 to £1,650 (bar); £1,725 by £75 to £2,175 per annum. Passages paid for appointee, wife and up to five children under 11 years, on appointment, annual overseas leave and termination. F.S.S.U. Children's, car and outfit allowances. Part-furnished accommodation at rent not exceeding 7.7 per cent of salary.

Detailed applications (six copies), naming three referees, by June 20, 1961, to the Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1, from whom further particulars may be obtained.

DEPARTMENT OF SCIENCE

KUMASI COLLEGE OF TECHNOLOGY
KUMASI, GHANA

Applications are invited for the following posts:

(1) Senior Lecturer and Lecturer in Mathematics.
(2) Lecturer in Chemistry.
(3) Lecturer in Botany.
(4) Lecturer in Zoology.
(5) Two Lecturers in Physics.

Qualifications:

Applicants should have an honours degree in any of the above subjects for which they are applying and be able to teach to the Advanced Level General Certificate of Education. The Lecturers in Mathematics should be able to teach mathematics to engineering students up to degree level. The Ghana Government intends to establish the College as an autonomous University in the very near future and teaching may then be to General degree level.

Contract appointments are normally for five years, and may be renewed. Three months' notice of resignation of an appointment is required on either side.

Contract Salary Scales
(Under Review)

Senior Lecturer in Mathematics:
£2,054 by £78 to £2,522.
Lecturers in Physics and Mathematics:
£1,110 by £60 to £1,950.
Lecturers in Chemistry, Botany and Zoology:
£1,063 15s. by £57 10s. to £1,868 15s.

Point of entry is related to previous experience.

In addition, children's allowances are paid up to a maximum of three, and a gratuity at the end of the contract at the rate of £150 for each year of satisfactory service.

The College is fully residential and members of staff are housed in pleasant modern bungalows on the spacious campus. Climate and living conditions are good.

Annual leave with three free return passages in four years is granted to members of staff and their families.

Application forms and an information Brochure may be obtained from the Assistant Registrar, Kumasi College of Technology, 29 Tavistock Square, London, W.C.1, who will also be glad to answer any questions either about the work of the School or about living conditions in Ghana.

BIOCHEMIST, NON-MEDICAL, BASIC grade, required as locum for period July 31 to September 2, 1961, at Group Laboratory, Epsom District Hospital. Salary, etc., as defined by the Whitley Council.—Applications to Group Secretary, Epsom District Hospital, Dorking Road, Epsom, Surrey, by May 31, 1961.

WOOL RESEARCH ORGANISATION OF NEW ZEALAND (INC.)
DIRECTOR

APPLICATIONS ARE INVITED FOR THE POSITION OF DIRECTOR, WOOL RESEARCH ORGANISATION OF NEW ZEALAND (INC.).

SALARY WILL BE IN THE RANGE OF £3,000-£3,500 STERLING DEPENDING ON QUALIFICATIONS AND EXPERIENCE. A SUBSIDISED SUPERANNUATION SCHEME IS AVAILABLE. REASONABLE TRAVELLING EXPENSES AND FARES TO NEW ZEALAND WILL BE PAID. IT IS DESIRED THAT THE APPOINTEE COMMENCE DUTY ABOUT APRIL 1962.

The newly formed Organisation is an autonomous body financed jointly by the New Zealand Wool Board and Government, and will have an annual income of £100,000. New laboratories are to be built adjacent to a university institution, Canterbury Agricultural College, Lincoln, near Christchurch.

The Director will be expected to plan and organise research into the physical, chemical and biological properties of wool and into other matters related to wool. He will be responsible to the Executive committee, and will be required to develop the Organisation from the initial planning of the laboratories, to the selection of staff, preparation of programmes of research, and dissemination of research findings.

Applicants should have a University degree in science or agriculture, and be experienced in organising and directing research preferably, although not necessarily, in wool science. Applications will be treated in strict confidence and should provide details of age, marital status, nationality, degrees held, names of referees, and full particulars of post graduate experience, and publications. Applications should be addressed to:

The Chairman,
Wool Research Organisation of N.Z. (Inc.).
Box 5115, Wellington, New Zealand.

Further particulars are available from the above, or either the Scientific Adviser, International Wool Secretariat, 18/20 Regent Street, London S.W.1, or the Director, Department of Science and Technology, The Wool Bureau Inc., 360 Lexington Avenue, New York 17, N.Y., U.S.A.

Applications close 31 July 1961

MIDDLESEX COUNTY COUNCIL
EDUCATION COMMITTEE
BRUNEL COLLEGE OF TECHNOLOGY
WOODLANDS AVENUE, ACTON, W.3
Principal: Dr. J. Topping, M.Sc., D.I.C., F.Inst.P.

RESEARCH ASSISTANT IN LIPID CHEMISTRY

Applications are invited for appointment to a Research Assistantship in Lipid Chemistry which has been established on a D.S.I.R. Award. Duties to commence on September 1, 1961, or as soon after as possible. The Research Assistantship is tenable for three years. Salary £870 rising to £905, subject to deductions for National Insurance and any superannuation scheme. The graduate appointed will be required to work on a project in Lipid Chemistry under the general direction of the Head of Department. Candidates should hold a good degree in chemistry or biochemistry and preferably have some research or industrial experience in the lipid field.

Further particulars and forms of application may be obtained from the Registrar, Brunel College of Technology, Woodlands Avenue, Acton, W.3, to whom they should be returned within fourteen days of the appearance of this advertisement.

C. E. GURR, M.Sc., Ph.D.,
Chief Education Officer.

SARAWAK
AGRICULTURAL OFFICER (DEVELOPMENT)

To administer an Agricultural Division with supervision of experiment stations, staff training and comprehensive advisory and supervisory duties. Candidates (male), between 25 to 40, must have a degree in agriculture or natural science with at least two years' postgraduate training or experience in tropical agriculture. Appointment on contract with gratuity of 12½ per cent of salary. Emoluments in range £1,344 to £2,324. Outfit allowance £60. Children's allowances. Education allowances. Free first-class sea passages on appointment and leave for officer, wife and up to three children under 18. Generous leave. Furnished accommodation usually available.

Apply, stating full name and giving brief particulars of qualifications and experience, to Director of Recruitment, Colonial Office, London, S.W.1. Quote BCD.63/24/014/C1.

RESEARCH CHEMIST

required to work in London on problems connected with drug synthesis. Must be well qualified and have research experience. Pharmaceutical qualifications and background an advantage.

Apply with full particulars to Professor E. E. Turner, F.R.S., Director of Research, Biorex Laboratories Ltd., 47/51 Exmouth Market, E.C.1, marking envelope "Research Chemist."

UNIVERSITY OF MELBOURNE
SENIOR LECTURER IN PHYSICS (R.A.A.F. ACADEMY)

Applications are invited for a position of Senior Lecturer in Physics at the Royal Australian Air Force Academy, which is affiliated with the University of Melbourne. Initial research will be related to studies of the atmosphere, radio research and cosmic radiation. Laboratories are situated at the University. The salary range is £A.2,480 to £A.2,950 per annum. Initial salary will be determined according to qualifications. Superannuation similar to F.S.S.U. in Great Britain will be provided.

Conditions of appointment may be obtained from the Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1. Applications close, in Australia and London, on June 30, 1961.

UNIVERSITY OF MELBOURNE
LECTURER IN PURE MATHEMATICS (R.A.A.F. ACADEMY)

Applications are invited for a position of Lecturer in Pure Mathematics at the Royal Australian Air Force Academy, which is affiliated with the University of Melbourne. The salary range is £A.1,660 to £A.2,360 per annum. Initial salary will be determined according to qualifications. Superannuation similar to F.S.S.U. in Great Britain will be provided.

Conditions of appointment may be obtained from the Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1. Applications close, in Australia and London, on June 30, 1961.

MIDDLESEX COUNTY COUNCIL
EDUCATION COMMITTEE
BRUNEL COLLEGE OF TECHNOLOGY
WOODLANDS AVENUE, ACTON, W.3
Principal: Dr. J. Topping, M.Sc., D.I.C., F.Inst.P.

DEPARTMENT OF MATHEMATICS

Applications are invited for an Assistant Grade B in Mathematics. Candidates should have a good degree, together with some research, industrial or teaching experience. A knowledge of some branch of modern applied mathematics such as operation research, numerical analysis or electronic computers would be an added advantage. The Department is responsible for teaching mathematics throughout the College, in Higher National Certificate, Diploma in Technology and postgraduate courses. There is a course leading to the Diploma in Technology in Mathematics. Salary: on scale £828 to £1,486 per annum, according to qualifications and previous experience.

Application forms from the Principal at the College, returnable within fourteen days of the advertisement.

C. E. GURR, M.Sc., Ph.D.,
Chief Education Officer.

CHELSEA COLLEGE OF SCIENCE AND TECHNOLOGY
MANRESA ROAD, LONDON, S.W.3
DEPARTMENT OF MATHEMATICS
RESEARCH ASSISTANTSHIPS

Applications are invited for two Research Assistantships in the Department. Candidates should possess a degree in Mathematics with first- or second-class honours. The successful applicants will prepare as internal students for a higher degree under a Recognized Teacher of the University of London. The posts are tenable for two years but may be extended to three years. The salary will be £400 per annum. The assistants may be required to demonstrate for a maximum of six hours weekly for which an additional payment will be made. Applicants should indicate the field of research or problem which they would like to investigate. In addition, they should give full particulars of their career and the names of two referees.

Applications should be received by the Clerk to the Governors not later than May 29, 1961.

NEWCASTLE UPON TYNE EDUCATION COMMITTEE

RUTHERFORD COLLEGE OF TECHNOLOGY

(Selected for Development into a College of Advanced Technology)

DEPARTMENT OF PHYSICS

The Governors invite applications for the undermentioned posts which arise due to the continued expansion of the Department and to the increased opportunities for research and similar undertakings. In all cases duties will commence on September 1, 1961. The Department is concerned almost exclusively with advanced courses leading to external degrees, professional membership, H.N.C., H.N.D., Diplomas in Technology, and with Postgraduate courses. Senior Lecturer (1 post): Candidates should have high academic qualifications together with teaching and, preferably, industrial and/or research experience. Lecturer (1 post): Candidates should hold, at least, a good honours degree, have had some teaching and, preferably, industrial and/or research experience. In each case an interest in one or more of the fields of Solid State Physics, Electronics, Nuclear Physics, and Optics would be an advantage. Salary will be in accordance with the Burnham Technical Report, viz.: Senior Lecturer, £1,550 by £50 to £1,750; Lecturer, £1,370 by £35 (final £40) to £1,550.

Further particulars and forms of application (returnable within 14 days) may be obtained from the Principal, Rutherford College of Technology, Northumberland Road, Newcastle upon Tyne, 1.

UNIVERSITY OF NOTTINGHAM

Applications are invited for the following appointments to begin January 1, 1962 (unless otherwise stated): Lecturer in Metallurgy, preferably with experience in industry and research. Lecturer in Chemical Engineering. Lecturers and Assistant Lecturers in the Mechanical Engineering Department in (a) Thermodynamics and Fluid Mechanics (October 1, 1961); (b) Mechanics and Materials. Assistant Lecturer in Medieval Economic History, preferably with knowledge of East European Languages and History. Salary scales: Lecturer, £1,050 to £1,850; Assistant Lecturer, £800 to £950; initial salary in each case according to qualifications and experience, plus membership of the Universities' Superannuation Scheme and children's allowance.

Conditions of appointment and form of application, to be returned by May 27, from the Registrar.

UNIVERSITY OF BIRMINGHAM

DEPARTMENT OF PHYSICS

Applications are invited for a Staff Fellowship in the Department of Physics. The Fellow will be expected to contribute substantially to the research programme in low-temperature physics being initiated within the Department, or to one of the existing programmes in nuclear physics and the physics of solids. Initial stipend within the scale: £1,050 by £50 to £1,400 by £75 to £1,550; £1,550 by £75 to £1,850. Appointment, tenable from October 1, 1961, will carry F.S.S.U. and family allowance benefits.

Applications (three copies), naming three referees, should be sent by May 24, 1961, to the Assistant Registrar (Science), the University, Edgbaston, Birmingham, 15, from whom further particulars may be obtained.

THE NATURE CONSERVANCY REQUIRE Scientific Officers for the following research programmes. Qualifications: good honours degree in subjects indicated, plus postgraduate ecological or relevant experience. Posts marked * for men only. (A) Invertebrate populations (base Wareham); Zoology; (B) *Coastal Physiography (Wareham); Botany; (C) Mountain Grasslands (Bangor); Botany; (D) Toxic Chemicals and Wild Life (Huntingdon); either Chemistry with Biology, Botany, or Agricultural Botany; (E) Conservation Research (Norwich); Zoology; (F) *Climatology (Aviemore); Geography, with Physics and Botany preferred; (G) either (i) Moorland Ecology or (ii) Plant Litter (Grange-over-Sands); Zoology (i); Chemistry or Biochemistry (ii). Starting salaries on scale £717 to £1,222 (for Post G only scale £1,302 to £1,604 may apply) according to qualifications and experience. Superannuation F.S.S.U.—Full details from the Nature Conservancy (E), 19 Belgrave Square, London, S.W.1, to whom applications (with appropriate No. of copies), giving full details of age, education, experience, National Service, natural history interests and other relevant information, with names/addresses of two referees for ability and personality, should be submitted by May 30, 1961.

ENGLISH CLAYS LOVERING POCHIN & COMPANY LTD.

St. Austell, Cornwall

A Company concerned with the production of China Clay (Kaolin, a basic raw material used in a wide variety of manufacturing Industries) and associated minerals. The producing pits, treatment plants and laboratories are situated mainly in the St. Austell area, near the South Cornish Coast.

THE RESEARCH DEPT.

has vacancies for Science graduates with or without post-graduate experience to work in the following sections:

Physical Chemistry: Two Chemists for work on the surface properties and general physical chemistry of clay minerals.

Chemistry: An Inorganic Chemist to work in a team engaged on the chemistry of minerals and associated problems.

Chemical Engineering: Preferably a young Chemical Engineer, or a Chemist, to join a team concerned with the development work on new processes for classifying, drying, mixing and mechanical handling.

Fuel Technology: A young Chemical Engineer or Chemist with a primary interest in Fuel Technology to study combustion problems and take part in development work on new processes.

Paint, Rubber and Plastics: A Senior Assistant for work on the application of the Company's products in the paint, rubber and plastics Industries and for Sales development.

Concrete and Building Materials: Head of Section concerned with development of China Clay by-products as raw materials for building.

Applications giving full career and personal particulars should be addressed to the Personnel Officer, 14 High Cross Street, St. Austell, Cornwall.

COLLEGE OF AERONAUTICS

ADVANCED STUDENTSHIPS IN MATERIALS SCIENCE AND ENGINEERING

A number of advanced studentships are available in the Department of Materials to enable Honours Graduates in Science or Engineering to enter one-year or two-year postgraduate courses in Materials Science and Engineering. These studentships are provided by certain well-known industrial organizations, and each selected candidate will be appointed to the staff of one of these organizations, by arrangement, on admission to the course, at the salary of £850 per annum. His tuition fee will also be paid. The course work comprises lectures and laboratory work, selected to meet the needs of the individual from a wide range of subjects dealing comprehensively with both metallic and non-metallic materials, and will include a research project under personal supervision. Successful completion of the course will lead to the award of the Diploma of the College of Aeronautics (D.C.Ae.) or the Diploma in Advanced Engineering (D.A.E.) according to the nature of the course undertaken. Those suitably qualified might be able to submit work for a higher degree.

Further information on the courses available and application forms may be obtained from the Warden, The College of Aeronautics, Cranfield, Bletchley, Bucks.

JOHN INNES INSTITUTE REQUIRE AN assistant with chemical background to help in biochemical investigations in potatoes in the Department of Potato Genetics. The appointment will be in the Assistant Experimental Officer grade, and a pass degree or equivalent qualification is required. Salary will be in the range £550 to £800 per annum, according to age and experience. Superannuation.—Applications, with the names of two referees, should be addressed to the Secretary, Bayfordbury, Hertford, not later than June 5, 1961.

UNIVERSITY OF THE WITWATERSRAND

JOHANNESBURG

DEPARTMENT OF PHYSICS

VACANCIES: RESEARCH FELLOW RESEARCH STUDENT

Applications are invited for the above temporary research posts. The successful applicants will be required to participate in the research of a small group working on problems of brittle fracture and on the influence of adsorbed substances on the strength of solids such as silica and tungsten carbide. Applicants for the Fellowship should hold the Ph.D. degree, and applicants for the Studentship should hold an M.Sc. or honours degree. The Fellowship is tenable for at least two years and may thereafter be renewed annually for a further two years. The Studentship will be tenable for the period of study for an M.Sc. or Ph.D. degree, subject to satisfactory progress being made. The salary attached to the Fellowship will be within the range R.3,060 to R.3,660 (£1,530 to £1,830) and the initial salary will be determined by the experience of the applicant. The salary in the case of the Studentship will be within the range R.1,680 to R.1,920 (£840 to £960) per annum for an M.Sc. graduate and for an honours graduate will be R.1,200 (£600) per annum (fixed). Passage to Johannesburg will be paid to a maximum of R.700 (£350) in the case of a married man and R.300 (£150) for a single person. Should the member of staff leave the service of the University within two years the money so advanced is refundable on a pro rata basis. Overseas return fares will be paid at half the above rate after three years and at the full rate after four years of service.

The successful applicant should assume duties as soon as possible. Applications in writing should be addressed to Professor F. R. N. Nabarro, c/o the Registrar, University of the Witwatersrand, Milner Park, Johannesburg, by not later than June 30, 1961.

UNIVERSITY OF NEW ENGLAND

ARMIDALE, NEW SOUTH WALES, AUSTRALIA

LABORATORY TECHNICIAN OR SENIOR LABORATORY TECHNICIAN IN BIOCHEMISTRY

Applications are invited for the above position and it is expected that applicants should have a good general background in Biochemical Laboratories. In the case of applicants outside Australia and New Zealand, I.S.T., I.M.L.T., or similar qualifications will be preferred. Duties will include assisting in both research and teaching. Weekly wage will be determined according to qualifications and experience within the following ranges: Laboratory Technicians, £A.18 9s. 3d. to £A.26 8s. 6d.; Senior Laboratory Technicians, £A.28 3s. 1d. to £A.30 5s. 5d. (current wage base £A.14 17s. per week for males.) Liberal recreation, sick and long service leave apply.

Applications, stating full details of qualifications and experience and the names of three people to whom reference about technical ability may be made, should be directed to the undersigned not later than May 31, 1961. Applicants outside Australia and New Zealand should forward an additional copy to the Secretary, the Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1, by the same date, who will arrange for the collection of referee's reports.

K. R. LONG,
Acting Registrar

University of New England,
Armidale, N.S.W., Australia.

ROYAL VETERINARY COLLEGE

(UNIVERSITY OF LONDON)

Applications are invited for appointment as Assistant Lecturer in Chemistry tenable in the Department of Physiology. Salary: £800 by £50 to £950 per annum, plus £60 per annum London allowance. Initial salary within the scale according to experience and qualifications. Superannuation benefits and children's allowances.

Application forms, obtainable from the Secretary, Royal Veterinary College, Royal College Street, London, N.W.1, should be returned not later than June 3, 1961.

UNIVERSITY OF READING

Applications are invited from persons with special qualifications in pure mathematics for Lectureship in Mathematics (Grade I or II). The appointment will date from September 30, 1961, or as soon thereafter as can be arranged.

Further particulars may be obtained from the Registrar (Room 22, O.R.B.), the University, Reading, by whom applications must be received not later than May 29, 1961.

EAST AFRICAN AGRICULTURE AND FORESTRY RESEARCH ORGANIZATION

MUNITALP RESEARCH FELLOWSHIP IN AGRICULTURAL METEOROLOGY

This Research Fellowship is available for work with the Munitalp Mobile Meteorological Laboratory, which will be centered partly at the Headquarters of this Organization at Muguga, Kenya, and partly at the Headquarters of the East African Meteorological Department at Nairobi. Applicants should preferably possess an honours degree in physics, and have had several years' postgraduate experience in physics or meteorology, and an interest in instrumentation would be an advantage. The value of the Fellowship is £1,750 per annum, and is tenable, in the first instance, for one year, but is renewable for a further two years by mutual agreement. Air passages to and from Nairobi will be provided for the successful applicant and his family. The Mobile Laboratory will be equipped with balloons (kytoons) to carry radiation recorders and other meteorological equipment, and will be used primarily to study the heat balance over land surfaces covered with different types of vegetation, under increasing severities of drought. Some of the work will be carried out over river catchments in which the complete water balance is being measured. The Fellow will work under the Head of the Physics Division of this Organization, which comprises three physicists, three experienced Scientific Assistants (Experimental Officers), a computing staff and other ancillary staff. The Fellow will be based at Muguga, which is 17 miles from Nairobi, nearly 7,000 ft. above sea level, and there are over 30 Research Officers on the Station.

Applicants are requested to write in the first instance to the Director, the East African Agriculture and Forestry Research Organization, P.O. Box 21, Kikuyu, Kenya. The Director, Dr. E. W. Russell, can be contacted from June to early September at 34 Hayward Road, Oxford.

BECHUANALAND PROTECTORATE

FORESTRY OFFICER

To organize and carry out a forest survey; to undertake research on regeneration; and to administer and supervise exploitation. Candidates must have a degree in forestry with postgraduate experience of forestry work, preferably in inventory work and general administration. Appointment on contract (gratuity terms with salary in scale £888 to £1,848. Gratuity 12½ per cent of aggregate salary. Furnished quarters at moderate rental. Free passages on appointment for officer, wife and dependent children under 21 years of age up to a maximum of five adult passages in all. Generous leave. Children's education allowances.

Apply, stating name in full and giving brief details of qualifications and experience, to Director of Recruitment, Colonial Office, London, S.W.1, quoting BCD.63/76/016.

SIR JOHN CASS COLLEGE

JEWRY STREET, LONDON, E.C.3
DEPARTMENT OF CHEMISTRY

The Governors invite applications for Lectureships in Physical and in Inorganic Chemistry. Applicants should have an honours degree in chemistry with postgraduate experience of academic or industrial research. For the Inorganic post applicants should have interest or experience in Radiochemistry and Analytical Chemistry. Salary £1,408 by £35 to £1,601.

Details and forms of application, which should be returned as soon as possible, may be obtained from the Secretary.

UNIVERSITY OF BELFAST

Applications are invited for a postdoctoral fellowship, salary £850, plus F.S.S.U., for work on the synthesis of organic compounds in the Department of Chemistry, the Queen's University of Belfast.

Applications, together with the names and addresses of two referees, should be sent to Professor H.B. Henbest by June 30, 1961.

APPLICATIONS ARE INVITED FOR A post of Junior Research Assistant in the Cytogenetics Group of the Paediatric Research Unit, Guy's Hospital Medical School. Applicants should possess a good honours degree in a biological subject, preferably with specialization in zoology or genetics, and will be expected to work in a group studying human chromosomes. Salary according to age, qualifications and experience.—Applications (two copies), to be sent to the Paediatric Research Unit, Guy's Hospital Medical School, London Bridge, S.E.1, to arrive not later than May 26, 1961.

C.A.V.

have a vacancy in their Research Department for a

GRADUATE IN PHYSICS

(or possibly Electrical Engineering)

Preference will be given to candidates with experience of *X*-ray diffraction techniques primarily as applied to the metallurgical field. A wide variety of problems arise in this work. Applicants should be versatile and able to devise and construct equipment as necessary.

Write in full (quoting Ref. F) to:

Staff Employment Officer,
C.A.V. Limited,
Warple Way, Acton, W.3.

CENTRAL ELECTRICITY GENERATING BOARD

BIOLOGIST

required at the Research Laboratories, Leatherhead, Surrey, to lead a Group investigating marine ecology around coastal power stations. An important part of the work will be to establish the relationship between the hydrography and the biology of the water.

Candidates should have a first- or second-class honours degree, preferably in zoology, or higher qualification, and some experience in this type of work. They should be prepared to spend a substantial part of their time on field work. Supporting staff is already available.

Salary will be fully commensurate with qualifications, ability and experience, and will be within the range £1,600 to £2,395 per annum.

Applications, stating age, qualifications, experience, present position and salary, to the Appointments Officer, 24/30 Holborn, London, E.C.1, by May 23. Envelopes should be marked "Confidential Ref. N/71."

UNIVERSITY OF QUEENSLAND

DEPARTMENT OF ANIMAL HUSBANDRY

The George Aitken Pastoral Research Trust Fellowship is offered in the field of bovine infertility. The specific aspect under investigation is a study of the relationship between anoestrus and protein intake. The Department of Animal Husbandry has been conducting field trials into the problems of infertility in central west Queensland for a number of years and the new appointee will be undertaking fuller investigations of an aspect presented by field evidence. In approved cases the work undertaken may be used towards a postgraduate degree. The salary will be within the range £A.1,250 to £A.1,600 according to qualifications.

Applications, stating age, qualifications, experience and other relevant information should be forwarded to the undersigned not later than May 27, 1961.

C. J. CONNELL,
Registrar.

University of Queensland,
Brisbane, Australia.

AUSTRALIAN NATIONAL UNIVERSITY

INSTITUTE OF ADVANCED STUDIES
DEPARTMENT OF GEOPHYSICS

A number of staff appointments are available in positions between the grades of Reader and Research Fellow. Appointments will be made preferably but not necessarily in the following branches of the subject: (i) classical geophysics including gravity and seismology; (ii) isotopic age determinations; (iii) palaeomagnetism; (iv) physics of high pressures; (v) structural geology or petrology. The department is engaged on work in seismology, palaeomagnetism, isotopic age determinations, deformation studies of rocks and minerals, phase equilibrium studies at high pressures and temperatures, spectro-chemistry and trace element analysis, meteorite studies, igneous and metamorphic petrology. It possesses a wide range of apparatus including high pressure equipment, mass spectrometers, an ARL X-ray microprobe and a full range of X-ray and optical equipment. Applicants may be appointed, according to qualifications, to the following posts: Reader, not less than £A.3,250; Senior Fellow, £A.3,000 to £A.3,550; Fellow, £A.2,120 to £A.3,000; Senior Research Fellow, £A.2,620 to £A.3,160; Research Fellow, £A.1,625 to £A.2,450.

Further particulars should be obtained from the Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1. Applications close, in Australia and London, on August 15, 1961.

UNIVERSITY OF CANTERBURY

CHRISTCHURCH, NEW ZEALAND
SENIOR LECTURER OR LECTURER IN PHYSICS

Applications are invited for the above-mentioned post. The successful candidate will be expected to take an active part in assisting, and extending the research being carried on in the Department, namely solid state studies, radio investigations of the upper atmosphere such as meteorite distribution, auroral zone detection and studies of the lower ionosphere. It is hoped to appoint someone who is prepared to work in the field of the solid state but applications are invited from persons suitably qualified and interested in any of the research topics mentioned. The salary for a Lecturer will be at the rate of £1,250 per annum, rising by annual increments of £75 to £1,700 per annum and for a Senior Lecturer at the rate of £1,750 per annum rising by annual increments to £2,000 per annum. Commencing salary will be in accordance with qualifications and experience. Approved fares to Christchurch will be allowed for an appointee, his wife and children. In addition actual removal expenses will be allowed within certain limits.

Further particulars and information as to the method of application may be obtained from the Secretary, Association of Universities of the British Commonwealth, 36 Gordon Square, London, W.C.1. Applications close, in New Zealand and London, on July 31, 1961.

UNIVERSITY OF MANCHESTER

Applications are invited for the post of Research Fellow in Chemical Crystallography in the Department of Chemistry in the Faculty of Science to undertake investigations into the structures of complex natural products of biological importance and novel structural interest. Excellent facilities are available for single crystal analysis and electronic computation. The Fellowship, valued at £875 per annum, will be available for one year from October 1, 1961, in the first instance, but renewal may be possible. Applicants should have had training in single crystal analysis and should possess a Ph.D. degree or have had at least three years' postgraduate experience.

Applications should be received not later than May 31, 1961, by the Registrar, University of Manchester, Manchester, 13, from whom further particulars and forms of application may be obtained.

MACAULAY INSTITUTE FOR SOIL RESEARCH

Applications are invited from graduates with first- or second-class honours in Mathematics with Statistics, or a Diploma in Statistics, for a vacancy in the Statistics Section. The duties are mainly concerned with the design and analysis of field experiments. The appointment will be made in the Scientific Officer Grade, starting salary within the range £738 to £1,222 per annum, according to age, qualifications and experience. Superannuation Scheme.

Forms of application may be obtained from the Secretary, the Macaulay Institute for Soil Research, Craigiebuckler, Aberdeen, to whom they should be returned before June 10, 1961.

AUSTRALIA
SENIOR BIOLOGIST
FISHERIES BRANCH, CHIEF SECRETARY'S DEPARTMENT, NEW SOUTH WALES

Applications are invited for appointment to the above position. Commencing salary between £A.2,290 per annum and £A.2,425 per annum, according to qualifications and experience. Qualifications: A degree in science from a recognized university with a major in zoology. Applicants must have undertaken original research preferably in aquatic fields since graduation, and published scientific papers. Duties: The successful applicant will be in charge of the Scientific Section of the Fisheries Branch and will be responsible for: (a) Planning and co-ordinating approved research programmes. (b) Supervision of the work of scientific staff and their briefing and training. (c) Checking reports and recommendations of scientific staff. (d) Carrying out original research work and advising the Superintendent of Fisheries on the management and development of the Fisheries. The successful applicant will be eligible, subject to passing a medical examination, to contribute to the State Superannuation Fund. Reasonable travelling and removal expenses will be paid to the successful applicant.

Form of application obtainable from the Office of the Agent-General for New South Wales, 56 Strand, London, W.C.2, with whom applications should be lodged by June 5, 1961.

UNIVERSITY COLLEGE OF GHANA

Applications are invited from suitably qualified candidates for appointment as Chief Technician in the Department of Zoology. Candidates must have adequate laboratory experience, and specialist experience of many of the following techniques: section cutting and staining; embalming of specimens for dissection; coloured latex injections; alizarin red preparations; preparation of osteological exhibits, culture media, stains, fixatives and chemical solutions; mounting of museum exhibits in perspex cases; maintenance of genetical stocks of *Drosophila*. The relevant Final City and Guilds Certificate or equivalent or higher qualification would be an advantage. The appointment is superannuable for Ghanaians on the salary scale £G.1,200 by £50 to £G.1,500, plus personal transport allowance, or on a four-year contract for non-Ghanaians. In the latter case the following payments will be made: 15 per cent contract addition, family allowance, outfit allowance, and a gratuity of £G.150 per annum. Passages on appointment, leave and completion of contract. Part-furnished accommodation at not more than 7.5 per cent of basic salary.

Applications (six copies), naming three referees, to be sent before June 5, 1961, to the Assistant Registrar (London), University College of Ghana, 15 Gordon Square, London, W.C.1, from whom further particulars may be obtained.

UNIVERSITY OF BELFAST

Applications are invited for a Research Fellowship in the Department of Chemistry of the Queen's University, Belfast, to develop methods for the study of adsorbed species on metals by infra-red spectroscopy, in collaboration with Dr. A. D. E. Pullin. The appointment will be for one year from October 1, 1961, but may be extended for a second year. Salary in the range £800 to £1,100 plus F.S.S.U. benefits. Applicants should have a research degree and preferably some experience of the operation and/or design of infra-red spectrophotometers.

Letters of application, including both details of scientific experience and interests and the names and addresses of two referees should reach Dr. A. D. E. Pullin, Department of Chemistry, Queen's University, Belfast, 7, by June 10, 1961.

THE HOSIERY AND ALLIED TRADES Research Association requires a Physicist of sufficient stature and experience to take charge of investigations of fundamental importance to the Hosiery Industry. This is a senior appointment, and is likely to appeal to a man who combines sound scientific knowledge with an imaginative outlook. Applicants should be between 25 and 35. They should have a good science degree and preferably experience of research. Experience of textiles would be an advantage but is not essential. Publication of research is encouraged. F.S.S.U. Salary will be commensurate with qualifications but will not be less than £1,400 per annum. The Association's premises are exceptionally well equipped and are ideally situated in pleasant surroundings in Nottingham.—Applications should be submitted in writing to the Secretary, H.A.T.R.A., Thorneywood, 7 Gregory Boulevard, Nottingham.

IMPERIAL CANCER RESEARCH FUND
DIVISION OF CHEMISTRY AND BIOCHEMISTRY

Applications are invited for the following appointments:

SCIENTIFIC OFFICER Grade III

Applicant should have research experience in biochemical techniques commonly used in metabolic investigations and should possess a Ph.D.

Initial salary within the range £1,050 by £100 to £1,250 per annum according to experience and qualifications plus £60 London allowance; F.S.S.U. benefits and family allowances.

SCIENTIFIC OFFICER Grade IV

Applicant would be required to assist in the synthesis of new organic compounds of potential physiological significance and should have recently obtained an honours degree in Chemistry. Full details of this and of any previous experience should be submitted.

Initial salary within the range £850 by £50 to £950 per annum plus £60 London allowance; F.S.S.U. benefits and family allowances.

SCIENTIFIC OFFICER Grade III or IV

Applicant should be a graduate or post-doctoral biochemist and would be required to work in the Section of Protein Chemistry.

Initial salary within the range £850 by £50 to £950 per annum, (Grade IV) or £1,050 by £100 to £1,250 per annum (Grade III) according to qualifications and experience, plus £60 London allowance; F.S.S.U. benefits and family allowances.

Applications, giving full particulars, should be addressed to: The Director, Imperial Cancer Research Fund, Lincoln's Inn Fields, London, W.C.2.

UNIVERSITY COLLEGE OF GHANA

Applications are invited in for (A) Lectureship or (B) Assistant Lectureship in Botany. Preference given to candidates with special interest in either general or biochemical plant physiology. Salary scales (under review): (A) £G.1,000 by £G.50 to £G.1,350 by £G.75 to £G.1,800; (B) £G.1,850 per annum. (B) £G.900; £G.950 per annum. Entry point according to qualifications and experience. F.S.S.U. Outfit, family and other allowances. Passages for appointee and family on appointment leave and normal termination. Part-furnished accommodation at a charge not exceeding 7.5 per cent of salary.

Detailed applications (six copies), naming three referees, by June 20, 1961, to Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1, from whom further particulars may be obtained.

UNIVERSITY COLLEGE
IBADAN, NIGERIA

Applications are invited for the Chair of Geography. Salary £3,000 per annum. Appointment to commence October, 1961, if possible. Passages paid for appointee, wife, and up to five children under 11 years, on appointment, annual overseas leave and termination. Children's, car and outfit allowances. F.S.S.U. Part-furnished accommodation at rent not exceeding 7.7 per cent of salary.

Detailed applications (eight copies), naming three referees, by June 20, 1961, to Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1, from whom further particulars may be obtained.

UNIVERSITY OF CAMBRIDGE
STATISTICIAN

Applications are invited for the post of Statistician to the Medical School. Applicants should be graduates in mathematics, mathematical statistics, or statistics. Experience with punched cards and electronic computers would be an asset. The appointment will be for five years from October 1, 1961, in the first instance. Salary, £1,250 by £50 to £1,400 by £75 to £1,800, according to experience.

Applications should be made in writing to Dr. G. A. Gresham, the Medical School, Tennis Court Road, Cambridge, not later than June 4, 1961.

LINCOLNSHIRE RADIOTHERAPY CENTRE
PHYSICS LABORATORY

Applications are invited for the post of Isotope Laboratory Technician in the Lincolnshire Radiotherapy Centre now being set up at St. George's Hospital, Lincoln. The work involved is the use of various types of Isotopes both on patients for diagnostic studies and therapeutic treatment and also experimental studies. Candidates should have a good school record with passes in physics and mathematics at O. or advanced level of the G.C.E. or O.N.C. in applied physics or inter-B.Sc. or equivalent qualifications. Salary scale at age 21, £570 to £710 per annum.

Applications, giving details of education, qualifications, and experience, together with the names of two referees, should be addressed to the Group Secretary, Lincoln No. 1 Hospital Management Committee, County Hospital, Lincoln, from whom further particulars can be obtained.

SOUTH-EAST ESSEX TECHNICAL COLLEGE
LONGBRIDGE ROAD, DAGENHAM

Required September, 1961: Assistant Lecturer (Grade "B") in Metallurgy. Applicants should have a degree or equivalent qualification, with teaching experience, and be able to teach to Higher National Certificate standard. Industrial experience is also desirable. Salary scale: £700 by £27 10s. to £1,150 per annum, plus London allowance (£38 or £51). In assessing the commencing salary, increments above the scale minimum will be allowed for approved teaching, research, or industrial experience and National Service. Additions to the salary scale will also be allowed for approved training and qualifications if in accordance with Burnham Further Education Report.

Application form and further particulars from the Clerk to the Governors.

BRITISH TECHNOLOGY INDEX

Applications are invited for the post of Editor of the *British Technology Index* which will be published monthly with an annual cumulation, commencing in January, 1962. The *Index* will analyse leading British periodicals in all branches of technology. Commencing salary within the range £1,500 to £1,700, contributory superannuation scheme. Duties to commence September 4, 1961. Applicants must have a knowledge of cataloguing and should have wide experience of technical information service in a technical library or possess a degree in science. The successful candidate will be required to set up the organization for preparing the *British Technology Index* and to plan the undertaking in detail, to act as editor after staff have been appointed, and will be responsible to the Council of the Library Association for all matters connected with the *Index*.

Applications, giving details of age, education and experience, together with the names of not more than three to whom reference may be made, should be sent to the Secretary, the Library Association, Chaucer House, Malet Place, London, W.C.1, by June 10, 1961.

UNIVERSITY COLLEGE OF GHANA

Applications are invited for (A) Lectureship or (B) Assistant Lectureship in Mathematics. Preference given to candidates with qualifications in applied mathematics. Salary scales (under review): (A) £G.1,000 by £G.50 to £G.1,350 by £G.75 to £G.1,800; £G.1,850 per annum. (B) £G.900; £G.950 per annum. Entry point according to qualifications and experience. F.S.S.U. Outfit, family and other allowances. Passages for appointee and family on appointment, leave and normal termination. Part-furnished accommodation at a charge not exceeding 7.5 per cent of salary.

Detailed applications (six copies), naming three referees, by June 25, 1961, to Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1, from whom further particulars may be obtained.

COUNTY OF ESSEX
SOUTH-EAST ESSEX TECHNICAL COLLEGE LONGBRIDGE ROAD, DAGENHAM

Applications are invited for appointment as a Research Assistant to carry out fundamental research in problems in organic or physical chemistry. Candidates should hold a good honours degree. Salary: £400 per annum, plus additional payment for part-time teaching.

Further details from the Clerk to the Governors at the College.

UNIVERSITY OF ST. ANDREWS

Applications are invited for an Assistantship in Organic, Physical or Inorganic Chemistry in the Department of Chemistry, Queen's College, Dundee, commencing October 1, 1961. The successful candidate will be given every encouragement to pursue research either on his own or in collaboration as he chooses. The Department is well equipped for most techniques, especially in the fields of kinetics and spectroscopy. Salary scale: £800 by £50 to £950. Appointment for three years, with possibility of renewal for fourth and final year. F.S.S.U. Family allowance. Grant towards furniture removal expenses.

Applications (six copies) containing the names of three referees, to the undernoted not later than June 14, 1961.

PATRICK CUMMING,
Joint Clerk to the University Court.
Queen's College, Dundee.

COUNTY BOROUGH OF STOCKPORT

STOCKPORT COLLEGE FOR FURTHER EDUCATION

Senior Lecturer in Science Department required to teach either Inorganic Chemistry or Physical Chemistry to the standard of Final B.Sc. and G.M.R.I.C. Part II. Candidates must be Honours Graduates of a British university. Duties to commence September 1, 1961. Burnham Technical Scale. Agreed conditions of tenure.

Further particulars and application forms from the Principal, Stockport College for Further Education, Wellington Road South, Stockport, to be returned by May 20, 1961.

UNIVERSITY OF HULL

DEPARTMENT OF MATHEMATICS
LECTURESHIP OR ASSISTANT LECTURESHIP IN APPLIED MATHEMATICS
LECTURESHIP OR ASSISTANT LECTURESHIP IN PURE OR APPLIED MATHEMATICS

Applications are invited for the above posts, tenable from October 1, 1961. Salary: Lecturer, £1,050 to £1,850 per annum; Assistant Lecturer, £800 to £950 per annum, with placing according to qualifications and experience; F.S.S.U. and family allowances.

Further particulars may be obtained from the Registrar, to whom applications (eight copies) should be submitted by June 5, 1961.

QUEEN MARY COLLEGE

(UNIVERSITY OF LONDON)

Applications are invited for two new posts of Graduate Research Assistants to participate in research on the mechanism and biochemistry of photosynthesis in the Department of Botany. Candidates should have an honours degree in botany, biochemistry, or chemistry. Salary £550 to £750, according to qualifications. The appointment is for one year in the first place with a possibility of renewal for a second year.

Application forms, obtainable from the Registrar, Queen Mary College, Mile End Road, E.1, should be returned by May 20, 1961.

UNIVERSITY OF DURHAM

KING'S COLLEGE, NEWCASTLE UPON TYNE, 1

The Council of King's College invite applications for a Lectureship in Applied Mathematics. The commencing salary will be determined at a suitable point on the Lecturers' Scale (£1,050 to £1,850) according to the age, qualifications and experience of the successful applicant. F.S.S.U. and child allowance.

Further particulars may be obtained from the Registrar and Secretary of King's College, to whom applications, together with the names and addresses of three persons to whom reference may be made, should be addressed so as to reach him not later than May 24, 1961.

ASSISTANT EXPERIMENTAL OFFICERS required by Research Institute (Animal Virus Diseases), Pirbright, Surrey: (1) For Biophysics Department. Work involves application of physical-chemical methods (electron-microscopy, electrophoresis, ultracentrifugation, etc.) to the study of foot-and-mouth disease and other animal virus diseases. Applicants over 22 should have H.N.C. or equivalent. (2) For work connected with production of attenuated strains of foot-and-mouth disease vaccines. Previous experience in virological techniques, preferably including tissue culture, essential. Applicants over 22 should have H.N.C. or F.I.M.L.T., preferably with virology as one subject. Salary dependent on age and qualifications in A.E.O. scale £625 at age 22 rising to £983. Houses available for married applicants. Superannuation scheme.—Applications to Secretary.

UNIVERSITY OF BRISTOL

Applications are invited for the following appointments in the Department of Geography. (1) Assistant Lectureship, tenable from October 1, 1961. Candidates will be preferred who have specialized in Economic Geography and have interests in agricultural or bio-geography. Salary within the scale £800 by £50 to £950 per annum. (2) Demonstratorship, tenable for one year in the first instance, renewable annually up to a maximum of three years. Salary £450 per annum.

Further particulars may be obtained from the Registrar, The University, Bristol, 8, to whom applications should be submitted by May 29, 1961.

UNIVERSITY OF LONDON KING'S COLLEGE

The College will require on October 1, 1961, an Assistant Lecturer in Physics.

Salary scale: £800 by £50 per annum to £950. A London allowance of £60, family allowances, and F.S.S.U. benefits will also be payable.

Facilities for research in either experimental or theoretical physics will be provided.

Conditions of appointment and application forms may be obtained from the Registrar, King's College, London, Strand, W.C.2; completed forms must reach him by May 31.

UNIVERSITY OF LONDON KING'S COLLEGE

The College will require on October 1, 1961, a Lecturer in Anatomy. Applicants should be graduates with medical qualifications.

Salary on the Junior scale: £950 by £50 to £1,150 per annum, plus a London allowance of £60, family allowances, and F.S.S.U. benefits.

Full conditions of appointment and application forms may be obtained from the Registrar, King's College, London, Strand, W.C.2; completed forms must reach him by May 31.

UNIVERSITY OF CAMBRIDGE

DEPARTMENT OF PHYSICS
CAVENDISH LABORATORY

Research Assistant is required for work in the metal physics group under the direction of Dr. P. B. Hirsch. Applicants should have some knowledge of metallurgical techniques. A university degree is not an essential qualification.

Applications should be submitted to the Secretary to the Department of Physics, Cavendish Laboratory, Free School Lane, Cambridge, by not later than June 22, 1961. The names of two referees who have a recent knowledge of the candidate's work should be given. Salary in the range £750 to £1,000, to be fixed in accordance with age and experience.

UNIVERSITY OF BELFAST

The Senate of the Queen's University of Belfast invites applications for a Senior Lectureship in Mechanical Engineering from October 1, 1961. Preference will be given to candidates who are interested in Strength of Materials. Salary range £1,700 to £2,275 plus contributory pension rights under the F.S.S.U. Initial placing on the salary scale will depend on qualifications and experience. Applications should be received by May 31, 1961.

Further particulars may be obtained from G. R. Cowie, M.A., LL.B., J.P., Secretary to the University.

BIRKBECK COLLEGE

(UNIVERSITY OF LONDON)

Applications are invited for the post of Assistant Lecturer in Botany. Salary scale £800 per annum by £50 to £950 per annum plus London allowance of £60 per annum. Candidates should obtain further particulars from the Secretary, Birkbeck College, Malet Street, W.C.1. Closing date for applications: May 29, 1961.

WOOLWICH POLYTECHNIC

LONDON, S.E.18

Principal: Harold Heywood, D.Sc., Ph.D., A.C.G.I., Wh.Sch., M.I.Mech.E., M.I.Chem.E.

Applications invited for a Lecturer in Physics from candidates with an honours degree. The Physics Department is a large one with courses for the B.Sc. General degree and B.Sc. Special degree in Physics (Internal) of the University of London. There are facilities for research. A knowledge of either X-ray Crystallography or of Nuclear Physics would be an advantage, but is not essential. Salary: £1,370 by £35 to £1,550, plus London allowance £38 to £51.

Particulars and application form from Clerk to Governors to be returned by May 27, 1961.

UNIVERSITY OF EDINBURGH

DEPARTMENT OF CHEMISTRY

Applications are invited for posts of Assistant Lecturers in the Department of Chemistry. Candidates should have particular interests in physical chemistry, inorganic chemistry or organic chemistry. Salary scale £800 by £50 to £950 per annum, with superannuation benefit and family allowance where applicable. Tenure is normally for three years. The successful candidates will be expected to take up duty on October 1, 1961.

Further particulars may be obtained from the undersigned with whom applications (six copies), giving the names of two referees, should be lodged not later than May 31, 1961.

CHARLES H. STEWART,
Secretary to the University.

UNIVERSITY OF BRITISH COLUMBIA

LOW TEMPERATURE PHYSICIST

The University invites applications for a vacancy in the Low Temperature Laboratory. The appointment will be made at the rank and salary appropriate to the training and experience of the applicant. The minimum salaries are $7,000 for Assistant Professor, $9,000 for Associate Professor and $12,000 for the rank of Professor. The laboratory is well equipped. The requirements are for a promising young man to take charge and direct a large part of the work.

For fuller information write to: G. M. Shrum, Head, Department of Physics, University of British Columbia, Vancouver 8, B.C.

UNIVERSITY OF MANCHESTER

Applications are invited from graduates in Physics, Chemistry, Metallurgy, Biology or Engineering for the post of Assistant Lecturer in Dental Materials. The person appointed will be concerned with work on the properties of dental materials. Applications would be considered from persons intending to graduate in 1961. There is wide scope for research in this field and ample facilities are available. Salary on a scale £800 to £950 per annum with membership of F.S.S.U. and Children's Allowance Scheme.

Applications should be sent not later than June 3, 1961, to the Registrar, the University, Manchester 13, from whom further particulars and forms of application may be obtained.

ASSISTANT LIBRARIAN REQUIRED FOR Central Technical Library and Information Service of a large expanding group of widely varying engineering companies in the Midland area. Applicants should be Chartered Librarians aged between 25/35 years, and preferably have industrial experience. The successful candidate will deal with the cataloguing, classifying (U.D.C.), reference work, correspondence, written inquiries and prepare reports. Experience of Staff control is required. Minimum starting salary £800 per annum; actual salary according to age and abilities; contributory pension scheme and usual large company facilities.—Please write, giving full details of personal background, experience and initial salary required to: Labour Officer, Messrs. Rubery Owen and Co. Ltd., P.O. Box 10, Darlaston, Wednesbury, S. Staffs.

A GRADUATE (IN MEDICINE OR Science) is required in the Bacteriology Department to take part in research work into the problem of staphylococcal virulence. Previous experience in isolation and estimation of toxins an advantage. Salary according to qualifications and experience, up to £1,500 per annum. Several other problems including the metabolism and antibiotic sensitivity and resistance of staphylococci are being studied in this Department and there may be further openings in these fields.—Applications, with details of qualifications and experience, and the names of two referees, should be sent to the Dean, Guy's Hospital Medical School, London Bridge, S.E.1.

UNIVERSITY OF DURHAM
KING'S COLLEGE, NEWCASTLE UPON TYNE, 1

The Council of King's College invite applications for a Lectureship in Pure Mathematics. The commencing salary will be determined at a suitable point on the Lecturers' Scale (£1,050 to £1,850) according to the age, qualifications and experience of the successful applicant. F.S.S.U. and child allowance.

Further particulars may be obtained from the Registrar and Secretary of King's College, to whom applications, together with the names and addresses of three persons to whom reference may be made, should be addressed so as to reach him not later than May 24, 1961.

CERAMIC RESEARCH
(LIAISON)

A young science graduate, preferably with two or three years experience in industry, is required to develop a new branch of an existing liaison service to the ceramic whiteware industry. Some travelling will be involved. An amiable personality and the capacity to organize are major prerequisites. Appointment will be in the Scientific Officer grade, qualifying for membership of F.S.S.U.

Applications, in writing, to the Director of Research, Dr. N. F. Astbury, The British Ceramic Research Association, Queens Road, Penkhull, Stoke-on-Trent.

DEPARTMENT OF AGRICULTURE AND FISHERIES FOR SCOTLAND

Two temporary unpensionable posts for Assistant Experimental Officers for research work in the field and the laboratory on potato black-leg disease. Qualifications: University degree in agriculture or pure science with main subject botany or bacteriology. Headquarters—Edinburgh. Age under 28. Salary: £598 (age 21) to £801 (age 26 and over); scale maximum £983.

Apply, giving full details of education, qualifications and experience (including ability to drive a car), also date of birth, to Establishment Officer, Room 73, Department of Agriculture and Fisheries for Scotland, St. Andrew's House, Edinburgh, 1, by May 31, 1961.

UNIVERSITY OF BIRMINGHAM
DEPARTMENT OF ZOOLOGY AND COMPARATIVE PHYSIOLOGY

Applications are invited for the post of graduate research assistant to the Professor of Zoology and Comparative Physiology. Candidates, preferably male, should have an honours degree in zoology or physiology. Applicants with pass degrees will be considered. Manipulative skill and interest in electrophysiology or electron-microscopy desirable. Salary, according to age and qualifications, on the scale: £425 by £25 to £675.

Applications, naming two referees, should be sent to the Assistant Registrar (Science), The University, Edgbaston, Birmingham, 15.

UNIVERSITY OF HULL

Applications are invited for appointment to Lectureships in Physics tenable from October 1, 1961. Salary: £1,050 to £1,850 per annum, with placing according to qualifications and experience; F.S.S.U. and family allowances.

Further particulars may be obtained from the undersigned to whom applications (six copies) should be submitted by May 31, 1961.

W. D. CRAIG,
Registrar.

MIDDLESEX HOSPITAL MEDICAL SCHOOL

Chief Technician required to take charge of technical establishment in new department dealing with medical applications of radioactive isotopes. Extensive experience of design and construction of electronic equipment essential. Some administrative experience desirable. Salary on scale £905 to £1,085 per annum, plus £40 London weighting.

Applications, giving full details to Secretary, Physics Department, Middlesex Hospital Medical School, Cleveland Street, W.1.

INSTITUTE OF DISEASES OF THE CHEST
(UNIVERSITY OF LONDON)

Applications are invited for a research worker of Ph.D. or M.Sc. status to work in the Biochemical research laboratory on problems of tissue respiration. Salary on university scale.

Applications addressed to: The Secretary, Institute of Diseases of the Chest, Brompton, London, S.W.3.

FEDERAL GOVERNMENT OF NIGERIA

VETERINARY RESEARCH OFFICER

Required by the Department of Veterinary Research, Vom, to undertake investigations and research into problems of animal disease in Nigeria, research into vaccines and supervision of technical staff engaged in vaccine production.

Candidates should be members of the Royal College of Veterinary Surgeons, or have a qualification recognized by that College.

Appointment on contract for one tour of 12 to 18 months in the first instance with prospect of extension. Salary according to qualifications and experience in the scale £1,434 to £2,196 per annum (including Inducement Addition) plus gratuity of £150 per annum on satisfactory completion of service. Outfit allowance of £60 on salaries below £1,741. Free passages for officer and wife. Liberal home leave on full pay. Quarters and income tax at low local rates. Children's allowances while separated.

Write for application forms and further particulars, stating briefly age, qualifications and experience, to the Recruitment Attaché, Office of the High Commissioner for Nigeria, Nigeria House, 9 Northumberland Avenue, London, W.C.2, quoting Z.3/1.

THE METAL BOX COMPANY LIMITED
RESEARCH DEPARTMENT, ACTON

require a

MALE OR FEMALE ASSISTANT

to work in the Microbiological Control Section. Facilities are available for further study and a person with G.C.E. advanced level in science subjects is desirable.

Please apply to:

Ref. CS 5
The Metal Box Co. Ltd.
Research Department, Kendal Avenue,
Westfields Road, Acton, W.3.

UNIVERSITY OF QUEENSLAND
RESEARCH ASSISTANT

Applications are invited for a position of Research Assistant in the Department of Biochemistry, University of Queensland. The appointee will be required to assist in fundamental studies of cancer metabolism and will have the opportunity to proceed to a higher degree. The appointment will be for two years at a salary of £A.1,350. Applicants should possess a university honours degree in science with biochemistry as a major subject or equivalent qualifications with research experience in biochemistry.

Applications, stating age, qualifications and experience, should be forwarded to the Registrar, University of Queensland, Brisbane, Queensland, Australia, before May 27, 1961.

SIR JOHN CASS COLLEGE
JEWRY STREET, LONDON, E.C.3
DEPARTMENT OF MATHEMATICS

The Governors invite applications for the appointment of Senior Lecturer in Mathematics. Applicants should have a higher degree in mathematics and be prepared to initiate research in some branch of applied mathematics. The salary will be on the scale £1,588 by £50 to £1,801.

Details and form of application may be obtained from the Secretary.

CHEMIST REQUIRED ON OR BEFORE August 1, by the Sports Turf Research Institute, Bingley, Yorkshire. Applicants should possess a degree, preferably in agricultural chemistry, and have postgraduate experience.—Further details obtainable from the Secretary.

BEDFORD COLLEGE
(UNIVERSITY OF LONDON)
REGENT'S PARK, N.W.1

The Council invites applications from honours graduates for the post of Assistant Lecturer in Chemistry, preferably with research experience and an interest in physical, physical inorganic, or physical organic chemistry. Vacant October 1, 1961; open to men and women. Salary scale £800 by £50 to £950, plus £60 per annum London allowance, with initial salary according to qualifications and experience. Closing date for applications: June 10, 1961.

Full particulars from the Secretary.

WYE COLLEGE
(UNIVERSITY OF LONDON)

Applications are invited for the posts of Assistant Lecturer in Zoology, salary scale £800 by £50 to £950, plus London allowance of £60, and Demonstrator in Zoology, salary £600 to £750. Superannuation under the F.S.S.U.

Further particulars from the Secretary, Wye College, nr. Ashford, Kent, to whom applications should be sent not later than May 31.

UNIVERSITY OF LONDON
READERSHIP IN MATHEMATICS AT WESTFIELD COLLEGE

The Senate invite applications for the Readership in Mathematics (Pure Mathematics) tenable at Westfield College (salary scale £1,675 to £1,975 to £2,425 a year, plus £80 London allowance).

Applications (10 copies) must be received not later than June 8, 1961, by the Academic Registrar, University of London, Senate House, W.C.1, from whom further particulars may be obtained.

NORWOOD TECHNICAL COLLEGE
KNIGHT'S HILL, S.E.27

Required for September, 1961: Assistant, Grade B, to teach Chemistry to G.C.E. Advanced and Degree level. Applicants should have teaching experience, a good degree or equivalent qualification. Salary range £738 to £1,486, commencing and maximum salary according to age, qualifications and experience. Appointment temporary in first instance, but permanency available if satisfactory.

Application forms from Principal, returnable within two weeks (enclose self-addressed foolscap envelope). Please quote (FE.3/N/1111/5).

RESEARCH ASSISTANT
(BIOCHEMIST)

Required for interesting post involving biochemical research into the causes of mental deficiency. Suitable for first- or second-class honours wishing to proceed to Ph.D. Good working conditions and ample scope for original work. Salary in the range of £650 to £1,150 (N.H.S. terms) according to qualifications and experience.

Apply, in the first instance, to Dr. B. W. Richards, St. Lawrence's Hospital, Caterham, Surrey.

UNIVERSITY OF BELFAST

The Senate of the Queen's University of Belfast invites applications for a Lectureship in Botany from October 1, 1961. Salary range £1,050 to £1,850 plus contributory pension rights under the F.S.S.U. Initial placing on the salary scale will depend on qualifications and experience. Applications should be received by May 31, 1961.

Further particulars may be obtained from G. R. Cowie, M.A., LL.B., J.P., Secretary, the Queen's University of Belfast, Belfast 7, Northern Ireland.

RESEARCH ASSISTANT WANTED AT Stoke Mandeville Hospital to help with current research on an immunological project connected with Rheumatoid Arthritis. The person appointed would work directly with the Pathologist undertaking the research. Salary £750 to £1,000 per annum according to qualifications.—Inquiries and applications, giving details of experience and the names of two referees, to the Pathologist, Stoke Mandeville Hospital, Aylesbury, Bucks.

SENIOR PHYSICIST REQUIRED IN RADIOtherapy Department. Applicants must be graduates in physics, preferably with honours and should normally have had five years experience of hospital physics or with radio-elements. The post offers development work on physical and instrumental problems. Salary £1,250 per annum rising to £1,700 plus £50 London weighting allowance.—Applications, stating age, experience, qualifications and names of two referees, to the Secretary, Lambeth Hospital, Brook Drive, S.E.11, as soon as possible.

NICHOLAS RESEARCH INSTITUTE

ALDERLEY EDGE

STATISTICIAN/TOXICOLOGIST

Graduate required with training in statistics, and physiology or pharmacology. The position is that of toxicologist at the Nicholas Research Institute Biological Laboratories at Alderley Edge, Cheshire, and the applicant will be expected to supervise toxicological testing of new drugs and also provide statistical services for the biological laboratories. Starting salary £1,000 to £1,500 per annum depending upon qualifications. The Nicholas Research Institute is part of the Aspro-Nicholas Group of companies.

Please apply in writing stating age, experience and qualifications to the Personnel Manager (R/E/2), Nicholas Research Institute Biological Laboratories, 225 Bath Road, Slough, Bucks.

UNIVERSITY OF ST. ANDREWS

RESEARCH ASSISTANT IN CRYOGENICS (LIQUID He^3)

Applications are invited for a Research Assistantship in very low temperature physics in the Department of Natural Philosophy, St. Salvator's College, to assist in the design and construction of liquid He^3 cryostats and refrigeration cycles. Experience in the handling of liquid He^4 is essential, and of He^3 of advantage but not essential. The post is part of a D.S.I.R. Research Project. Salary £1,000 per annum for three years; for a suitably experienced person the salary may be higher for a shorter period. F.S.S.U. benefits if successful applicant is already within that scheme, but not otherwise.

Applications, with names of two referees, should be lodged with the Joint Clerk to the University Court, College Gate, St. Andrews, not later than June 15, 1961.

RESEARCH AND DEVELOPMENT ESTABLISHMENT

OF

AERONAUTICAL AND GENERAL INSTRUMENTS LTD.,
105/107 LANSDOWNE ROAD,
CROYDON

REQUIRE:

Senior Electronics Engineers who should be qualified at least to Higher National Certificate level and/or have a wide and varied experience of present day electronic technique.

All applications in the first instance to be in writing to the above address.

ROWETT RESEARCH INSTITUTE

Assistant Experimental Officer required in the Department of Applied Nutrition to work mainly on amino acid analyses of proteins by the "Spinco" Analyser. Candidates should have the Higher National Certificate in Chemistry or equivalent qualification. Experience of or interest in instrumentation would be of advantage. Salary scale (men or women) £450–10s. (age 18) to £957. A superannuation scheme is in force.

Applications, stating date of birth with particulars of qualifications and experience, together with the names of referees, should be sent to the Secretary, Rowett Research Institute, Bucksburn, Aberdeen.

GLASSHOUSE CROPS RESEARCH INSTITUTE requires Scientific Officer for plant breeding section, primarily for research on mushroom spawn and breeding for disease resistance in higher plants. Appointment initially in Scientific Officer scale (£717 to £1,222), according to age and experience; superannuation under F.S.S.U. Candidates should have honours degree in appropriate subject, and preferably postgraduate experience in genetics, particularly of micro-organisms, but students with suitable basic training graduating this year may be considered.—Further details available from Secretary to the Institute, Worthing Road, Rustington, Littlehampton, Sussex, to whom applications, with names of three referees, should be submitted before May 31, 1961.

GRADUATE CHEMIST REQUIRED, WITH several years' industrial experience, to perform and supervise inorganic analysis. A knowledge of modern physical methods including ion exchange and chromatographic techniques is desirable. The work will entail investigations and research programmes connected with conventional and nuclear power plant.—Applications, stating age and experience, should be sent to the Superintendent, Research Department, Babcock and Wilcox Ltd., High Street, Renfrew.

CLARKSON COLLEGE OF TECHNOLOGY

PHYSICS DEPARTMENT

POTSDAM, NEW YORK, U.S.A.

Applications are invited for a faculty appointment effective September 1, 1961. Salary and rank will depend upon academic qualifications and experience of appointee. Applicant should have a doctor's degree in physics and a strong interest in teaching at all levels from first year college through graduate. Participation in research will be expected. First preference will be given to theoretical physicist interested in solid state, second to experimentalist in same field.

Applications, giving full particulars of education and experience, and listing the names of three references, should be sent to Professor T. S. Renzema at the above address.

IMPERIAL CANCER RESEARCH FUND

Junior Biochemist required for new virus project. Experience in tracer techniques an advantage. Salary according to age, qualifications and experience.

Applications to Dr. R. J. C. Harris, Imperial Cancer Research Fund, Mill Hill, London, N.W.7.

UNIVERSITY OF LONDON KING'S COLLEGE

The College will require on October 1, 1961, or as soon as possible thereafter, a Lecturer in Electrical Engineering. Applicants should have a good honours degree in electrical engineering. Preference may be given to an applicant with special interests in electrical machinery.

Salary scale: £1,050 by £50 per annum to £1,400 then by £75 per annum to £1,625; then subject to review by £75 to £1,850. A London allowance of £60, family allowances, and F.S.S.U. benefits will also be payable.

Conditions of appointment and application forms may be obtained from the Registrar, King's College London, Strand, W.C.2. Completed forms must reach him by June 7.

INSTITUTE FOR NUTRITION RESEARCH

RUSCHLIKON-ZÜRICH

seeks qualified laboratory technician with biochemical background; experience in protein chemistry and some knowledge of German is of advantage but not necessary. Salary in accordance with age and experience.

The post is available from August 15, and applications, stating the date of birth, qualifications and experience, together with references, should be forwarded not later than June 30 to the Director (Institut für Ernährungsforschung, Seestrasse 72, Rüschlikon-Zürich, Switzerland).

MAKERERE UNIVERSITY COLLEGE

EAST AFRICA

Applications are invited for Lectureship in Physical Chemistry. Salary scale £1,350 to £2,151 per annum. Entry point determined by qualifications and experience. F.S.S.U. Child allowance £50 per annum per child (maximum £150 per annum). Passages for appointee and family (up to five adult passages) on appointment, termination and leave (three months every 21 months). Rent according to quarters provided £45 to £84 per annum (including basic furniture).

Detailed applications (six copies), naming three referees, by June 22, 1961, to Secretary, Inter-University Council for Higher Education Overseas, 29 Woburn Square, London, W.C.1, from whom further particulars may be obtained.

VACANCIES EXIST FOR POSTGRADUATE research in the chemistry and biochemistry of micro-organisms. Students will be eligible for registration for the Ph.D. (internal) degree of the University of London. Research grants may be available in October for students who attain a good honours degree in chemistry or biochemistry. Applications should be submitted as soon as possible by candidates who have already graduated or who will be sitting their final examinations before August, 1961.—Apply in writing to the Professor of Biochemistry, London School of Hygiene and Tropical Medicine, Keppel Street, W.C.1.

UNIVERSITY COLLEGE OF THE WEST INDIES

Applications are invited for post of Demonstrator or Assistant in sub-Department of Pharmacology. Duties include demonstration and some teaching for the M.B., B.S. degrees of London University and research. Initial appointment one year. Salary: medically qualified £1,000 per annum, non-medical in range £500 to £900 per annum, depending on qualifications and experience. Unfurnished accommodation at rental of 10 per cent of basic salary. Return passages for appointee only.

Detailed applications (four copies), naming three referees, by June 28, 1961, to Secretary, Senate Committee on Colleges Overseas in Special Relation, University of London, Senate House, London, W.C.1, from whom further particulars may be obtained.

DALHOUSIE UNIVERSITY

HALIFAX, NOVA SCOTIA, CANADA

Applications are invited for a position in the Department of Mathematics.

The initial salary will be from $7,000 to $9,000, depending on the qualifications of the applicant, who should write, including a short curriculum vitae, to Dean Hicks, Dalhousie University.

A SCIENCE GRADUATE (CHEMISTRY and botany) with an interest in horticulture is required to carry out research on the composition of fruit varieties. The appointment is permanent and prospects are excellent. Personal qualities are more important than high academic standard.—Applications should be addressed to the Director, Fruit and Vegetable Canning and Quick Freezing Research Association, Chipping Campden, Glos, giving full personal details and stating salary required.

LEVY WEST LABORATORIES OF Wembley require a graduate for work on very pure inorganic chemicals. Some physical leanings useful. Outside experience helpful but not essential. Salary according to qualifications.—Write or telephone WEMbley 1088.

WELL ESTABLISHED CHEMICAL MANUfacturers have vacancy for qualified Research Chemist age 28 to 30, preferably with some industrial experience in organic chemicals and synthetic resins. Modern laboratories, five-day week and contributory pension scheme. Salary according to age and qualifications £1,200 to £1,400.—Apply Managing Director, Synthite Limited, Ryders Green, West Bromwich.

AGRICULTURAL RESEARCH COUNCIL require an honours graduate in chemistry or biochemistry at the Low Temperature Research Station, Cambridge, for a period of about three years to investigate the constituents of animal connective tissue together with related enzyme systems. Good knowledge of general chemistry and an interest in protein chemistry essential. Subjects for study are of type generally suitable for submission as dissertation for a higher degree. Salary in the range £738 to £1,222, according to qualifications and experience.—Applications, with names of two referees, to the Director of the Station, Downing Street, Cambridge.

GRANTS & SCHOLARSHIPS

UNIVERSITY OF CAMBRIDGE

Applications are invited for two Research Grants tenable at the Department of Colloid Science from October 1, 1961, for theoretical and experimental work on particle interaction in colloidal systems. Successful candidates would be expected to become registered research students and to study for higher degrees in the University.

Further details from the Secretary, Department of Colloid Science, Free School Lane, Cambridge.

ROYAL HOLLOWAY COLLEGE

(UNIVERSITY OF LONDON)

POSTGRADUATE STUDENTSHIPS 1961-62

The Council of Royal Holloway College will shortly award a certain number of Postgraduate Studentships in Science, open to men and women graduates of any university. Successful candidates, if eligible in other respects, will be recommended for Studentships of the Department of Scientific and Industrial Research. Particulars may be obtained from the Registrar.

Applications should be forwarded to the Principal, Royal Holloway College, Englefield Green, Surrey, as soon as possible, the closing date for the receipt of applications being July 1, 1961.

UNIVERSITY OF LEEDS

THE HOULDSWORTH SCHOOL OF APPLIED SCIENCE

CHARLES BROTHERTON SCHOLARSHIP IN CHEMICAL ENGINEERING, GAS ENGINEERING AND FUEL SCIENCE

Applications are invited for the above scholarship of the annual value of £125 a year, provided to enable promising students to follow an honours course in chemical engineering, gas engineering and fuel science in the Houldsworth School of Applied Science. In cases where the candidates reach the required standard, the scholarship may be supplemented by a State Scholarship awarded by the Ministry of Education. The scholarship is open to applicants of British nationality from school or industry, who are qualified to enter upon a degree course.

Applications should reach the Registrar, from whom further particulars and forms of application may be obtained not later than June 30, 1961.

UNIVERSITY OF LEEDS

THE HOULDSWORTH SCHOOL OF APPLIED SCIENCE

Applications are invited for the following entrance awards, tenable in the Houldsworth School of Applied Science:

William Cartwright Holmes Scholarship in Gas Engineering and Fuel Science, tenable in the Houldsworth School of Applied Science. The present value of the scholarship is £140 a year and it is tenable for one, two or three years, with power of extension for a fourth year.

Corbet and Henry Woodall Scholarship in Gas Engineering and Fuel Science, tenable in the Houldsworth School of Applied Science of the value of £135 a year. It is tenable for three years with power of extension for a fourth year.

The Scholarships are intended to enable a student to take a course of study at the University in preparation for a responsible position as a Gas Engineer. They may be supplemented by the award of a supplemental State Scholarship by the Ministry of Education where a candidate reaches the required standard.

Further particulars and forms of application may be obtained from the Registrar, the University, Leeds 2, to whom applications should be made before June 30, 1961.

UNIVERSITY OF LEEDS

OPPENHEIMER RESEARCH SCHOLARSHIPS IN AFRICAN GEOLOGY

Applications are invited from suitably qualified graduates in geology, for Oppenheimer Research Scholarships tenable in the Research Institute of African Geology. The work, which will lead to a Ph.D., will be done partly in Africa and partly in Leeds. The scholarships, which are awarded for a period of three years, are worth £600 a year, together with certain field allowances and provision for fares to and from Africa each year.

Applications, together with the names of two referees, should be submitted on or before June 15, to Professor W. Q. Kennedy, Department of Geology, the University, Leeds, 2, from whom further particulars may be obtained.

UNIVERSITY OF MANCHESTER

METROPOLITAN-VICKERS POSTGRADUATE BURSARIES

Applications are invited from honours graduates in Engineering, Physics, or Mathematics for a number of postgraduate bursaries of a value normally within the range of £350 to £700 per annum, although this maximum may be exceeded in certain cases. The bursaries are tenable for research and advanced study in the Departments of Electrical and Mechanical Engineering in the Faculty of Science or of Technology.

Applications, naming the Department and Faculty in which the candidate proposes to work, should be sent not later than July 1, 1961, to the Registrar, the University, Manchester, 13, from whom further particulars and forms of application may be obtained.

HUMAN GENETICS

DARWIN RESEARCH FELLOWSHIP

Applications are invited for a Darwin Research Fellowship in the field of human population genetics with special reference to the range of human variability. The Fellowship, preferably full-time and up to three years in duration, would carry a salary in the range £800 to £2,000 and is open to persons suitably qualified in science or medicine.

Application should be made by June 15 (although a later application might be considered) to the Eugenics Society, 69 Eccleston Square, London, S.W.1, from which further information may be obtained.

UNIVERSITY OF LONDON. APPLICATION is invited for the Laura de Saliceto Studentship (part-time) for the advancement of Cancer Research, value £150 a year for not less than two years. Applications should reach the University not later than June 1, 1961.—Further particulars may be obtained from the Secretary to the Scholarships Committee, University of London, Senate House, London, W.C.1.

LECTURES AND COURSES

BATTERSEA COLLEGE OF TECHNOLOGY

(A COLLEGE OF ADVANCED TECHNOLOGY)

CRYSTALLOGRAPHY SECTION

A postgraduate course leading to the examinations for the degree of M.Sc. in Crystallography of the University of London or the Postgraduate Diploma of the College (D.C.T.(Batt.)), will begin on October 2, 1961. Full-time or part-time students are accommodated, and in certain circumstances scholarships are available.

For further details apply to Dr. D. Lewis, Head of the Crystallography Section, Battersea College of Technology, Battersea Park Road, London, S.W.11.

TRAINING PROGRAMME IN STEROID BIOCHEMISTRY

Applications now being received for one year advanced training in the theory and technology of steroid biochemistry to start September, 1961. Recent graduates with M.D. or Ph.D. degree will receive stipend of $5,500. Sponsored by National Cancer Institute, N.I.H.

Apply Dr. Frank Ungar, Department of Physiological Chemistry, The Medical School, University of Minnesota, Minneapolis 14.

BRADFORD INSTITUTE OF TECHNOLOGY

DEPARTMENT OF CHEMICAL ENGINEERING

A Short Course on

"REFRIGERATION IN THE CHEMICAL INDUSTRY"

will be held on Thursday and Friday, June 29 and 30, 1961. Course fee: £1 10s.

Further details and forms of application may be obtained from the Head of Department of Chemical Engineering, Bradford Institute of Technology, Bradford 7.

UNIVERSITY OF LONDON: A LECTURE entitled "The Analysis of Cell Reproduction" will be delivered by Professor D. Mazia (California) at 5.30 p.m. on May 18 at the School of Pharmacy, 29-39 Brunswick Square, W.C.1. Admission free, without ticket.—James Henderson, Academic Registrar.

BATTERSEA COLLEGE OF TECHNOLOGY, London, S.W.11. The Spectroscopy Section has organized a Summer School on "Colour Technology (II)," to be held June 19-24. Fee 12 guineas, or 10 guineas if revision work (first day) is not required.—Further details and enrolment forms available from the Secretary (Spectroscopy Courses).

U.K.A.E.A.

HARWELL REACTOR SCHOOL

STANDARD COURSE NO. 23

To be held from September 18, 1961, to February 2, 1962. Fee £250 exclusive of accommodation. This Course has been revised to include the latest information of interest to engineers and physicists whose main concern is reactor design or operation. It now includes a period of nine weeks at a College of Advanced Technology in addition to the ten weeks of more advanced study at A.E.R.E. Harwell and A.E.E. Winfrith.

CONTROL AND INSTRUMENTATION OF REACTORS COURSE NO. 7

To be held at Durley Hall, Bournemouth, from September 25 to October 6, 1961, inclusive. Fee £52 10s. exclusive of accommodation. It is intended for those who have a direct interest in the control and instrumentation of nuclear reactors, and participants are assumed to have some knowledge of the basic principles of these subjects.

SENIOR TECHNICAL EXECUTIVES' COURSE NO. 13

To be held from September 11 to 22, 1961, inclusive, of which the first week will be at the Reactor School, Harwell, and the second at Durley Hall, Bournemouth. Fee £52 10s. exclusive of accommodation. An appreciation course for senior executives which covers the background to reactor physics and engineering including simple experiments. Applications of nuclear engineering are illustrated by visits to A.E.R.E. Harwell and A.E.E. Winfrith and a nuclear power station.

Application forms and further details for these courses can be obtained from:

The Manager, Reactor School,
A.E.R.E., Harwell,
Berks.

BATTERSEA COLLEGE OF TECHNOLOGY

LONDON, S.W.11

The fifth One-Year Full-time Course in "Control Engineering" will commence on October 2, 1961. The course is recognized by the D.S.I.R. for the tenure of Advanced Course Studentships and holders of a second-class honours degree may qualify for the award of a Studentship.

Applications should be submitted to the Head of the Electrical Engineering Department, Battersea College of Technology, Battersea Park Road, London, S.W.11.

KENT EDUCATION COMMITTEE

KENT SUMMER SCHOOL, FOLKESTONE

35TH ANNUAL SESSION

July 29 to August 12, 1961

Principal: Professor Ben Morris, B.Sc., Ed.B. (Director, University of Bristol Institute of Education)

The main aim of this Summer School will be to give specialist teachers in secondary schools an opportunity of widening their knowledge of their subject. Courses will be offered in the following subjects: English, Divinity, Natural History and Ecology, Mathematics, Chemistry, Physical Education (men and women), and Art and Crafts (Fabric Printing and Weaving). Inclusive fees: for Resident Students: Kent, £7 5s.; non-Kent, £27 10s. For Day Students (exclusive of meals): Kent, £3 17s. 6d.; non-Kent, £14.

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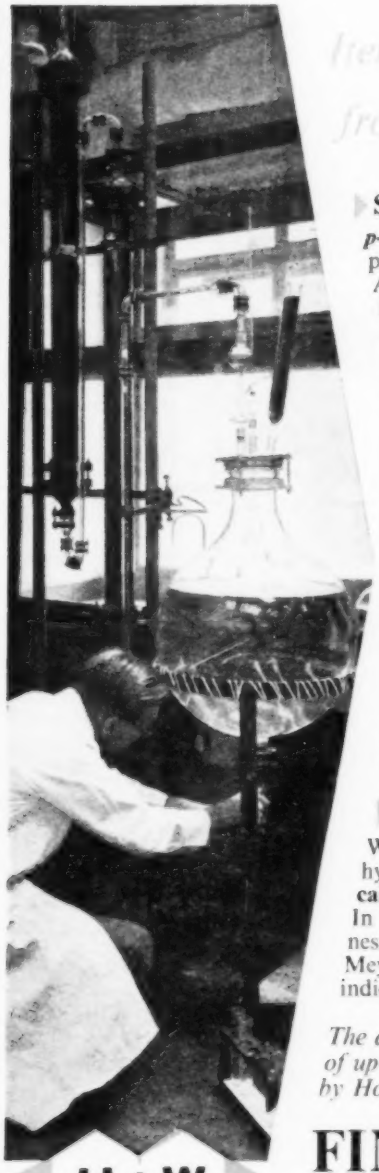
Incidental Information

No. 14

Items of interest from our laboratory notebooks



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We make the reagent and can supply from stock. A suitable Lovibond Comparator for this test is obtainable from Baird & Tatlock (London) Ltd.

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Our **1-(2-pyridylazo)-2-naphthol** (**PAN** for short), already known as a metal indicator for complexometric titrations, (H. Flaschka and H. Abdine, *Mikrochim. Acta*, 770 (1956); *Anal. Abs.*, **4**, 794 (1957)) is now described as a spectrophotometric reagent of high specificity for **uranium** (H. Shibata, *Anal. Chim. Acta*, **22**, 479 (1960)).

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We also make **di-(*o*-hydroxyphenylimino) ethane** (glyoxal-bis-(2-hydroxy anil)), a specific colour reagent for microgram quantities of **calcium** in solution.

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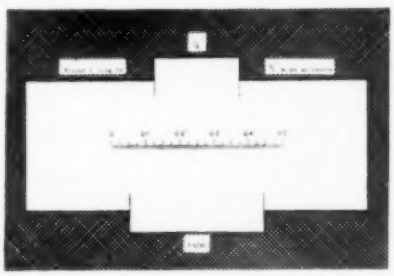

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